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STUDIES ON SOME FLAGELLATA.

BY E. PENARD.

While studying Infusoria, in the neighborhood of Geneva, Switzerland, during the years 1914 to 1918, I often happened to meet with particularly interesting Flagellata, and sometimes interrupted my ordinary work to study some of these organisms. The results of these investigations appear of sufficient interest for publication, and it is a particular pleasure for me to make them known here. The species to be discussed are: *Pteridomonas scherffeli* Lemm., *Dimorpha tetramastix* sp. n., *Dimorpha monomastix* sp. n. *Bicoeca exilis* sp. n., *Histiona campanula* sp. n., *Salpingoeca polygonatum* sp. n., *Salpingoeca lepidula* sp. n., *Chrysamoeba radians* Klebs, *Chrysopyxis bipes* Stein, *Hyalobryon ramosum* Lauterborn, *Cryptomonas ovata* Ehrenberg, *Astasia mobilis* (Rehberg) Alexieeff, *Euglena pseudomermis* sp. n., *Trentonia flagellata* Stokes.

Pteridomonas scherffeli Lemm Plate V, figs. 1–3.

Though the description of the genus *Pteridomonas* dates back as far as 1890 (Penard, 28), the first concise diagnosis which we possess is due to Lemmermann (24), and I can do no better than to reproduce it here, at least in its most important points:

> *Pterdomonas* Petard. Cells naked, mostly stalked; 1 flagellum, and rarely, besides, 2 slowly vibrating accessory flagella. At the base of the flagellum, a crown of fine radiating pseudopodia. One or two contractile vacuoles. Nucles with distrinct caryosom. Multiplication unknown. Food taken by the pseudomopodia, or by means of special food-vacuoles. Movement freely rotating, swinging, or by sudden leaps backward, due to the contraction of the stalk.

As for *Pteridomonas scherffeli*, which was found by Scherffel in Hungary, then by Lemmermann at Bremen, Germany, its specific characters are indicated as follows: "Cells generally longer than broad, in optical longitudinal section rounded with 5–6 angles. Stalk long and fine. Nucleus near the base of the flagellum. Besides the anterior crown of pseudopodia, some lateral radiating pseudopodia."

This *Pteridomonas scherffeli* was found in great quantities in the month of September last (1918), among the decaying leaves that filled the bottom of a pond, at the Ariana Park, near Geneva.

I was then enabled to study this very interesting organism at some length, and should like to discuss here some of the most important points of its structure.

Regarding the identity of my specimens: Lemmermann speaks of "cells longer than broad, in optical longitudinal section rounded with 5–6 angles," while in the Ariana specimens the breadth of the body was normally at least equal, and very often superior, to the length; and at the same time the anterior part was broader than the posterior (Plate V, fig. 1); in optical longitudinal section the form, rather than "rounded with 5–6 angles," was that of a top with a truncated point. In short, this Ariana form was but in a very general manner in accordance with Lemmermann's description; but as this very small organism is difficult to study, and is perhaps subject to great variation in its shape, it seems best at present to consider the two forms as representing the same organism.

Pteridomonas in form, as we just said, is more or less that of a top; the size is about 8–9μ, often rather less, and rarely more. In the anterior part of the body we find, right and left, two small contractile vesicles, and also, in the posterior part, two other vaculoes, but of a special nature; the body, in fact, seems to be hollowed out to form two small depressions, one at the left and the other at the right, with a clear space or vacuole in the center; they hardly look like contractile vesicles, especially since these depressions are sometimes wanting, yet sometimes I have seen the vacuoles suddenly collapsing, as contractile vesicles generally do. The flagellum, whose length is equal to three or four times that of the body, is relatively strong, and easily seen. From the middle of the anterior part of the body, which is itself somewhat flattened or even excavated, this flagellum points directly upwards during the first part of its course, then curves right or left, and when vibrating becomes hardly visible. At its point of fixation it is provided with a basal granule, very small and mostly quite invisible, but which I could detect once on a gradually and strongly compressed specimen.

In Lemmermann's diagnosis we noted the following lines: "One flagellum, and rarely besides, two slowly vibrating accessory flagella. At the base of the flagellum, a crown of fine radiating pseudopodia." We must treat at some length of these rather important organs.

The central flagellum is normally surrounded by a crown of very fine threads, perhaps about one dozen in number, quite rigid and motionless, which point straight into the surrounding liquid, at an angle of about 45° from the central flagellum. These threads, which have generally been described as pseudopodia, and indeed certainly appear to be such; very often seem to start, not from a middle region quite near the flagellum, but further away from the middle, even from near the extreme border of the body, like an anterior crown above the equatorial girdle of setae which we shall soon speak of. But it only appears so, and in fact one can easily see, on specimens favorably oriented for such observations, that all the threads of the crown have their origin in the immediate vicinity of the central flagellum. Sometimes, left and right of the flagellum are seen two much smaller flagella, slowly undulating, and quite independent from the big central one. These two small accessory flagella, whose meaning has not yet been understood, seem to have been a great puzzle for those few observers who noticed them; but more protracted observations, with adequate material, would have allowed of more certain conclusions. In fact, there are not always two accessory flagella, but sometimes one or two, or more; or, on a specimen where none was at first detected, suddenly there may be seen to appear a crown of small flagella, which begin vibrating, and after a very short time slowly retract, getting thicker the more they shorten—as if an axial thread were retracting into the body while the enveloping protoplasm accumulated outside—and finally collapsing each of them into a small roundish pearl. But after a time, each of these little pearls rapidly lengthens again into a long, fine thread, that becomes mixed among the other threads or "pseudopodia" which had remained expanded and rigid.

These observations, which have been repeated several times, seem to justify the following statement: The threads which compose the anterior crown are not pseudopodia, but flagella, which are capable of changing rapidly in their appearance and function; straight and rigid as a rule, they may for a time appear as flagella, with slow, flexuous undulations. However, these accessory flagella widely differ in their nature from the central one, which is a true flagellum. They are also completely independent from each other, one of them, for instance, is seen to vibrate while its next neighbor remains stiff; or some developing into a vibrating crown of flagella, while others persist in the shape of a rigid external crown of pseudopodia (Plate V, fig. 2).

As to the function of these special elements, for which we might use the term "flagellopodia," in their rigid, pseudopodian state, they must be considered as prehensile organs, stopping on their way very small organisms or food particles and bringing them towards the body; on two different occasions, for instance, I have seen a small green particle caught by one of the rigid filaments, and the thread curving over it, then slowly retracting and finally depositing it on the surface of the body. This process is, however, not the only one possible, and much more frequently the small food particle, without coming into contact with the extended threads, directly gets to the surface of the body, where a big vacuole is immediately formed around it; but it remains certain that these rigid filaments, twice or three times as long as the body, form together something like a net, and as such are of very evident importance. But besides these functions as prehensile organs, such as are observed in the pseudopodial state, could not we imagine functions of a different nature, due to the flagelliform state of these particular threads? When describing for the first time, in 1890, the genus *Pteridomonas*, I felt confident I had seen the small pearls at the base of the central flagellum suddenly unroll and fling the little animal backward; the flagellopodia, in fact, were to be compared to the frond of a fern and not to a compact shapeless body. But Scherffel, in 1901, proposed another explanation; —he looks upon the threads as true pseudopodia, quite incapable of passing into another condition; the two smaller flagella, which are at times seen left and right of the central flagellum, "are in no way two pseudopodia which have taken to an undulating condition, as Penard thinks. Their function is unknown"; and a little further on he adds: "The jumps are due to the energetic and sudden contractions of the fixed stalk." Much later, in 1904, after observing some specimens of *Pteridomonas* from the Lake of Geneva, I had myself abandoned my former explanation to adopt the Scherffel theory, and today, after a much more thorough examination, I feel confident the rolling and unrolling explanation was a mistake; but is the sudden jerk backward due really to a contraction of the pedicle?

In *Pteridomonas*, the body is protracted behind into a thread, of extraordinary thinness, like the thread of a spider; and this thread or pedicle, four, six, or as much as eight times as long as the body, firmly holds the animalcule to the substratum. In such an attached condition, the little organism is generally seen fishing, with its central flagellum actively swinging, and the threads of the anterior crown rigid, unmoved, and expanded at their full length, but from time to time the animalcule suddenly disappears from sight, and rapidly comes back, a jerk has thrown him to one side but the fixed pedicle prevented any definitive removal. This posterior thread is thus very important in enabling, for instance, the animalcule to escape the contact of an enemy and yet remain in the same region; but it is hardly to be expected that the pedicle might do more than attach the body, and be comparable to the retractile pedicle of a *Vorticella.* Its whole appearance is that of a very thin, thread-like secretion, where no structure is to be found; and indeed, facts may be cited which go against the retractile explanation:

Looking at a fixed *Pteridomonas* for a sufficient time, a moment at last comes when the pedicle suddenly loses its hold, the crown of pseudopodia retracts, and the little animal swims rapidly away, while the detached posterior thread or pedicle trails behind. In such a state of rapid locomotion, with pedicle trailing or sometimes with no pedicle at all (owing to its having been ruptured or having collapsed), when coming into contact with some Infusorian or any other obstacle in its way, the animalcule may suddenly jerk backward, just as it did when attached.

Under these conditions it seems hardly possible that the stalk might be the cause of the sudden jerk; and we must look for another explanation.

The most evident distinctive character between *Pter. scherffeli* and *Pter. pulex* is the possession, by the first of these species, of lateral filiform appendages, which are wanting in the second. Lemmermann speaks of these appendages as follows: "Besides the anterior crown of pseudopodia, lateral radiating pseudopodia are found." Neither Scherffel nor Lemmermann speak of these "pseudopodia" as being regularly distributed, and the first of these observers speaks of both anterior crown and lateral appendages in the following terms: "The fine setose cilia, which radiate mostly from the anterior part of the body, around the flagellum, *but sometimes also start, in a smaller number, from the sides of the body,* are true pseudopodia." Both these authors, in fact, seem to consider these lateral appendages as identical with those of the anterior crown, and at the same time as irregularly located on the sides of the body. But the reality is, that these particular threads, six in number (or perhaps eight ?), are inserted in a single equatorial line, or girdle, which itself is, in most of the individuals, indicated by a slight groove that divides the body into two parts, superior and inferior. These threads are extraordinarily thin, are straight and rigid, and are twice or even three times as long as the body; they never are converted into flagella, and never retract into separate pearls, and are in no way comparable to the constituent parts of the anterior crown—they rather seem to be identical with those setae with which some Infusoria (*e. g., Mesodinium pulex*) are provided. Now in these Infusoria the setae always work as a jumping organ; and why should their function not be the same in *Pteridomonas?* There is a fact, indeed, which seems to contradict such a theory,— *Pteridomonas pulex,* which has no such setae, jumps just as well as *Pt. scherffeli* which possesses setae, but . . . it is not quite certain that *Pter. pulex* is unprovided with setae. None have been seen, it is true, but they are so extremely indistinct, in *Pter. scherffeli,* that during the first days of my observations I had no idea it was not *Pter. pulex* I had to do with, and only later on the mistake was found.[1]

It is, then, to that aequatorial girdle of setae that I should be disposed today to attribute the jerks of *Pteridomonas,* but the question yet remains in doubt.

When speaking of the anterior crown, we saw that each of its "flagellopodia" is liable to collapse into a small globular body, and that all of these small pearls constitute then a sessile ring around the central flagellum. Now it is rather interesting to notice that this very sort of ring sometimes is found also at the posterior extremity (Plate V, fig. 1). In that particular locality, the Ariana Park, which furnished practically all my material, this posterior crown existed in one out of five individuals, being sometimes well developed, more often more or less reduced in the number of the "pearls", but none of these pearls ever could be seen to develop into a thread; they were always in the retracted state, and very likely were unable to change in any way.

As to the meaning of that posterior crown of pearls, at first sight it seems that it would indicate the anterior part of a new individual, to be formed by an act of division which must then prove transverse. In spite of the fact that a transverse division would be very exceptional in a Flagellate, I was inclined to consider it as very probable here, till meeting one particular specimen which certainly was dividing (Plate V, fig. 3). It was much broader than long, with two flagella, a nucleus elongated transversally to the vertical axis of the body, and two posterior threads, each with some pearls around its base. This specimen showed the beginning of a *longitudinal* division. We must then conclude that the meaning of the posterior crown is not yet understood.

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[1] The first description of *Pter. pulex* is dated 1890. At that time I had at my disposal only a small, student's microscope, and practically the aequatorial setae must have been invisible. Besides, *Pter. pulex* hardly ever seems to have been studied afterwards. Scherffel and Lemmermann had *Pter. scherffeli* under observation, and though some other observers cite *Pter. pulex,* I do not know of any who studied it.

Such are the observations which I could make on *Pteridomonas*, a very small and delicate organism, but one whose structure proves very particularly interesting; and in my opinion the principal interest attaches to the special threads or "flagellopodia," whose functions are double, now pseudopodia, now flagella. Scherffelie, 1911, whilst treating of *Chrysopyxis bipes* and its pseudopodia, observes that the existence of food-capturing flagella has never been proved in any of the Protozoa; but in *Pteridomonas* we find something quite exceptional: the food is caught by pseudopodia, the undulating movements are due to flagella, but these very flagella were pseudopodia some minutes before, and next moment will be so again.

Dimorpha tetramastix sp. n. Plate V, figs. 4–6.

"Cells with distinct periplast, free-swimming. Pseudopodia numerous, with axial threads that radiate from a central granule situated inside the body. Two flagella, originating in the neighborhood of the central granule. Nuculus with distinct caryosome. One to several contractile vesicles. . . ."

Such is the diagnosis given by Lemmermann (in Pascher's "Süsswasser-Flora," p. 51) of the genus *Dimorpha*, which Gruber (14) created in 1881 for a small Protozoan whose characters are at the same time those of a Heliozoon and a Flagellate.

Dimorpha mutans is the only species that has been described; however, one other has been at least indicated. Blochmann (3), in 1894, after treating of *Dimorpha mutans*, concludes in the following terms: "I myself found, in the summer of 1885 in a pond of the Schwetzinger Garden, another form, of which I unhappily took only a few incomplete sketches. It differed from *D. mutans* in the possession of four flagella, and also in the fact that it always swam about with fully expanded pseudopodia."

In the month of July last, and later in August and September, I found in the same pond of the Ariana Park a species of *Dimorpha* provided with four flagella. Is it the same as that mentioned by Blochmann? Possibly so, but, as we shall see later, it is not certain. Is it even a *Dimorpha?* According to the first diagnosis of the genus, which mentions two flagella, it is not; but, like Blochmann, I should rather insist on the fact of the resemblance with an Heliozoon than on the possession of one, two or more flagella.

Dimorpha is indeed now a Heliozoon, and now a Flagellate. Let us first consider our *Dim. tetramastix* in the Heliozoon, that is, in the resting state (Plate V, fig. 4):

The spherical body, 30 to 33μ in diameter (much larger than *Dim. mutans*) is naked, but not smooth, owing to the presence of numerous granules of a relatively large size, some of which project a little outside the surface. Two layers, in fact, may be distinguished in the plasma, the ectoplasm with the above-mentioned granular bodies, and the clearer endoplasm; but the difference between the two layers is here much less distinct than in typical Heliozoa, those for instance which constitute the group of the Chalarothoraca. Around the body, and radiating in every direction, but less numerous anteriorly, are the pseudopodia, straight and very fine, whose length attains to 1½ to 2 times the diameter of the body. Just as in Heliozoa, they are covered with small granulations, but are even smaller here; and besides, they have sometimes a peculiar appearance, which does not seem to have been observed in Heliozoa as a rule: as long as they have not attained to their entire development, their tip is blunt, truncated at a right angle to the longitudinal axis; but when fully extended, they are pointed; one might suppose that in the first case the layer of viscous plasma which covers the axial thread did not extend further than the axial thread itself, and in the second case it had gone farther, lengthening to a point. Axial filaments exist here indeed, as in typical Heliozoa, and in favorable conditions they may be seen to penetrate the plasma and converge towards a central granule, which itself is easily distinguishable, and with carmine stains quickly. But this "central granule" is in fact never quite central but lies at a greater or less distance from the center toward the anterior extremity.

This central granule is itself surrounded by the special apparatus which Gruber (14), Blochmann (3), Schouteden (31) have already described in *Dimorpha mutans*, but which is here of a rather different nature, consisting of very small, yellow, spherical grains, which are set around the central granule like numerous radiating chaplets, owing, very likely, to their being attached to the axial filaments which converge towards the centre. All together, they build up a kind of spherical cage, quite characteristic of *Dimorpha tetramastix* and allowing one easily to distinguish this species from any Heliozoon in the vicinity.

Food is caught by the pseudopodia, and in a very characteristic manner. On four or five different occasions I observed the capture, and always it was the same small Infusorian, *Balanitozoon agile*, which suddenly came into contact with one or several of the pseudopodia; these then tightened around their captive, and at the same time a very thin film or velum came creeping over the pseudopodia, lengthened into a cup, and finally closed behind the Infusorian. This latter, which had been suddenly killed as by some special poison, became more and more elongated, then was cut into two parts, the first of which entered the body of the *Dimorpha;* the second fragment then divided again, and finally each of the separated portions got into the *Dimorpha* and quickly rounded itself into a small pellet. Our *Dimorpha* seemed in fact to feed exclusively on this little Infusorian, and proved excessively gluttonous. Three minutes are sufficient for the conversion of a living *Balanitozoon* into a number of pellets inside the *Dimorpha,* and this latter, which measured at first say about 30μ in diameter, increases to 40μ in diameter after the capture.

We must now consider the flagella somewhat at length. In a *Dimorpha* favorably oriented for good observation, one can distinguish in the body an anterior part, slightly flattened, and even sometimes depressed to form a shallow median cup, and from that cup arise four very fine threads, hardly visible, about twice the length of the body; they appear and suddenly disappear, and when lost are very difficult to trace again, owing to their extraordinary tenuity. Two of them are often seen to cross each other at a short distance from their base, which fact is due to their being in pairs, one of the pairs originating left and the other right of a central point of fixation (Plate V, fig. 4).

Gruber could not ascertain the presence of a nucleus, at least in the living animalcule, but says he easily demonstrated it with reagents. Blochmann, however, and afterwards Schouteden (31), each gave a rather complete description of what they considers a the nuclear apparatus, and it is interesting to quote what the latter of these observers writes upon the subject: "The nucleus is already visible in the living animal, more or less distinctly according to the individuals. But it is easy to make it appear by killing the organism with a solution of picric acid, which colors the nucleus yellow. One sees then that chormatin is united in a thick mass which in optical section appears like a crescent with more or less rounded or obtuse points; really this mass constitutes something like a hemisphere (or segment of a sphere, smaller or sometimes larger,) hollow but with a thick wall.

"Inside that peculiar mass, in the cavity which it surrounds, one sees the axial threads of the pseudopodia, which pass through the body, to converge toward the same point, namely to a shining, very distinct granule, which is indeed already visible, with some attention, in the living animal. In this same place, besides, is the insertion of the flagella, which diverge from the common point inside the plasma and come out of the body in two distinctly separated regions . . . . Blochmann was rather puzzled by the fact that the axial threads of the pseudopodia thus converge *through the nucleus* toward a common point. He does not think probable that there could be there a continuous axis which would pierce through the nuclear substance. And yet, according to my observations, the complicated apparatus of *Dimorpha* must indeed be thus explained. The axial threads which are seen converging run completely through the chromatic mass, pierce it in fact, and nevertheless they are absolutely continuous and homogeneous."

These observations are interesting, in so far as they show the flagella to be inserted quite near the central granule; but both Blochmann and Schouteden are certainly wrong when they consider as a nucleus that particular apparatus which we have seen in *Dimorpha tetramastix* consists in radiating lines of pearls. In reality, the true nucleus, generally concealed by the accumulation of granules and food-particles, easily comes to view by means of a slight compression of the animal. It quite conforms in character to an ordinary Heliozoan nucleus, being rather large, homogeneous, variable in its shape, and always somewhat excentric in position.[2] There are several contractile vesicles, variable in number, but so obscured by the mass of granules and food-particles that fill the external layers of the body, that one hardly sees them.

Such is the Heliozoan form of *Dimorpha tetramastix;* but if we keep the animalcule under observation for a while, after five, ten, twenty minutes or more, we can see its transformation into the flagellate state. Suddenly, and without any apparent motive, the pseudopodia are seen to retract, at the same time as the body lengthens, and the central granule with the surrounding chaplets of pearls moves forward; and after a single second of time, or at most two, the Heliozoan has disappeared, and instead we have a true Flagellate (Plate V, fig. 5), more or less lengthene, with an anterior depression from which arise the two pairs of flagella. Something, however, is frequently left of the pseudopodia, in the form of lateral or, more often, posterior appendages, more or less covered with granules (Plate V, fig. 6); and some of the axial threads are still visible, trailing inside the body, behind the central granule which is now quite near the anterior extremity.

It is possible only under particularly favorable conditions to get a clear view of the phenomenon of the mechanism of that sudden transformation, but I was able to observe it several times and with sufficient clearness to be able to explain the facts. The pseudopodia, at the same time that they retract and shorten, move backward while the central granule is brought forward; this displacement of the pseudopodia is, in fact, to be compared to the shutting of an umbrella, whose ribs are brought down and collapse together. As for the cause of the shutting, it must very likely be sought in internal movements inside the plasma, which result in an accumulation of clear pure ectoplasm at the anterior extremity; if we can imagine a current coming from behind and pushing the central granule forward along the longitudinal axis of the body, and at the same time lateral backward currents pushing the axial rods downward, we can get an idea of the transformation.

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[2] Blochmann, in his figure of *D. mutans* in the Heliozoan state, has in fact represented the nucleus, but as a "particle of food."

As it is, the Flagellate, which now measures about 37µ in length, *i. e.*, 7µ more than in the Heliozoan shape, immediately takes to swimming, rapidly drawn forward by its flagella. As already noted, some of the pseudopodia, or rather some of the axial threads with their covering of granulations, are very often seen trailing laterally or behind, but I never could see, as Blochmann said of his unidentified *Dimorpha*, the animalcule swimming with completely and normally expanded pseudopodia.

Swimming never lasts very long; the most trifling cause is sufficient to instigate reversion to the Heliozoan type; the animalcule is seen to stop, and suddenly the pseudopodia make their appearance, lengthening at the same time as they are brought forward, while the central granule moves downward toward the center of the body The same kind of phenomenon has taken place, which we compared to the shutting of an umbrella, but it is now the opening, and it has been even more rapid.[3] This transformation from the Flagellate state to the Heliozoan type is brought about by anything that might be supposed to indicate trouble or danger—a small Infusorian in the vicinity, a current of water, etc.—while the inverse, the transformation from the Heliozoan into a Flagellate, is much more difficult to produce; a strong light, however, which provokes the displacement of the animalcule toward a cooler region, is often successful. Very probably, the Heliozoan form is the form of defense, the pseudopodia firmly affixing the body to resist currents, and at the same time it is the fishing-form, allowing small organisms to be caught by the pseudopodia.

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[3] This reversion is still more shortened by the action of a chemical reagent; in one particular case, for instance, I have seen the transformation entirely effected in the half of a second, under the influence of a very weak current of glycerin and carmine.

Must we consider *Dimorpha* as a Heliozoon or as a Flagellate? As a Flagellate, of course, since it is an understood fact that all organisms are to be considered as such, which possess flagella in the adult state; but the affinities with Heliozoa are so great that, in fact, we might look upon this organism as a Heliozoon also. In some of the representatives of this latter group, for instance, *Clathrulina*, an embryonic stage is known, where the little being is in possession of flagella; and one might be entitled to ask if *Dimorpha* could not be a member of the same group, in which the flagella persist up to the adult state?

In this connection I should like to relate a rather curious coincidence: in the same locality there was living, in still much greater abundance, a genuine Heliozoon, of a yet undetermined type, and whose likeness to *Dimorpha tetramastix* was such that only the possession of the characteristic pearl apparatus and the non-existence of a flagellated state allowed an easy distinction. At the same time, there were occasions when I never could detect any trace of flagella in otherwise typical Dimorphas, and sometimes also the pearl apparatus was hardly to be seen. *Dimorpha* might be supposed to lose finally its flagella, as well as the pearl apparatus which in some unknown way might be in relation with them? But, leaving that supposition aside, what more beautiful example of mimicry could we imagine?

Dimorpha monomastix sp. n. Plate V, figs. 7, 8.

The preceding species was characterized by the presence of four flagella; now we find only one. *Dimorpha monomastix* was found in the same locality, and in a much greater abundance, but its small size makes it rather more difficult to study.

The little organism is about 10μ in diameter, transparent and colorless; in a superficial region are seen dispersed all sorts of granules and food-particles, surrounding an indistinctly delimited clearer central region or endoplasm, which contains the nucleus. This latter is spherical, with a distinct nuclear membrane and a large central caryosome. Very likely this endoplasmic region also contains a central granule, which, however, I did not see. There are several contractile vesicles, all very small, except one that might be considered as the principal one, the others being accessory. All around the body are the pseudopodia, of the typical Heliozoan type, but relatively short, measuring about 1½ times the diameter of the body. The flagellum, always single, is of a moderate length also, about the same as that of the pseudopodia, but strong and thick, much thicker, in fact, than in *Dimorpha tetramastix*. However, it is extremely transparent, and there is some difficulty in distinguishing it; a moderate enlargement, indeed, which allows of a very small diaphragm, is better for its detection than very strong magnification.

During the Heliozoan state of rest, the flagellum remains quiet, and nearly always with a peculiar shape, somewhat like a figure 8 (Plate V, fig. 7); from time to time, however, it is seen slowly to unroll, and to perform one or two slight movements, soon coming at rest again.

This Heliozoan type is the less frequent form, and the animalcule never keeps it for a very long time. After a few minutes of observation, the swinging of the flagellum grows quicker and more frequent, and suddenly the whole body seems to be subjected to a disturbance, a rupture of its internal equilibrium. It collapses, in fact, and one might take it to be disorganized and dead; but after hardly one or two seconds have passed, the whole mass, as by enchantment, lengthens into a fusiform body, having anteriorly a quickly-moving flagellum.

The little flagellated being (Plate V, fig. 8), which from 10μ in the Heliozoan state has now increased to 18μ in length, and even sometimes more, has the appearance of a fish, broader at the anterior part, and drawn posteriorly into an often very thin and long extremity, which seems to result from the fact that the axial threads have joined together and trail behind. The nucleus, very distinct, has come forward, very near the anterior extremity, and is seen surrounded by very small, hyaline granulations. The flagellum comes straight from a slight excavation in the middle of the anterior, rounded extremity. Several small contractile vesicles are visible, one of them larger, near the posterior end of the body.

The animalcule goes straight forward in a rapid course and swims for a very long time, for hours probably; then suddenly it stops, the flagellum becomes quiet and takes the characteristic figure 8 shape; again a commotion, which shakes the whole body, and rays are seen to protrude from every part of the surface. In two seconds, the Flagellate has turned into a Heliozoon.

The transformation is effected here in the same way as we saw it in *Dimorpha tetramastix*, but is much less evident to the eye.

Bicoeca exilis sp. n. Plate V, figs, 9–13.

This elegant little Flagellate was found, in February, 1918, rather abundant in one of the aquaria of the Zoological Laboratory of the University. Most generally it was attached to the threads of a small *Nostoc*, either solitary or with one or two companions, but never in big colonies.

The little capsule (Plate V, fig. 9), very thin and colorless, is 8μ in length; ovoid or somewhat fusiform in shape, pointed behind and opened anteriorly in a large aperture, at right angle to the longitudinal axis. This opening, however, is rather variable in breadth, according not only to the individuals, but also to the orientation of the animal; the capsule, in fact, is a little compressed, more so at the opening than anywhere else, and at the same time proves somewhat elastic; when the animalcule protrudes for half its length out of the shell, it presses against the sides and makes the opening rounder, and when suddenly retracting the edges of the aperture contract. The capsule is affixed to the substratum by means of a very thin pedicle, a simple colorless filament, whose length is exceedingly variable, and is commonly twice or three times as great as that of the capsule, sometimes even more. Frequently it is seen to terminate at the point of fixation in a small bluish knob or cushion, and from the fact that the volume of the knob is in inverse proportion to the length of the pedicle, one can infer that it furnishes the material for the lengthening of this thread. This growth of the pedicle, however, must be very quick, and reaches its limit a very few moments after the fixation of the animalcule; such is, at any rate, the inference that might be drawn from the fact that specimens with very short stalks (Plate V, figs. 9, 12), kept under observation for ten, twelve and twenty-four hours, never showed during that time any lengthening of their stems. Very likely the length of the pedicle is in some relation with the conditions of the medium; for instance, in some material which had been kept a long time since it was collected, I once found all the individuals short-stalked (Plate V, fig. 12). The animalcule hardly fills half of the capsule. In its perfectly expanded state, it is pear-shaped. At the anterior extremity and on one side is seen projecting a protoplasmic appendage which looks at first sight like a very short pseudopodium; but in reality the ectoplasmic layer developed here in a collar-like expansion, hardly conspicuous on one side but very extended on the other, and whose function is to hold fast the small particles of food that the flagellum has thrown down. The flagellum, which starts from the anterior depression, is strong and very distinct, and of very remarkable length, attaining five or even six times the length of the body. When the animalcule is quietly fishing, with its body half protruded from the shell (Plate V, fig. 9), the flagellum is seen pointing directly upwards, and hardly seems to move, but sometimes is only distinguishable up to the half of its length, the tip only vibrating. The animalcule does not, as a rule, remain very long in the extended state; it suddenly retracts inside the shell and becomes ovoid in shape, but for a very short time only, soon expanding again. The behavior of the flagellum is then very interesting to follow: when the animalcule retracts, the flagellum coils in a perfectly regular manner, so as to simulate a coil of rope with four or five turns, which stands upright over the retracted body (Plate V, fig. 11); then as soon as the animalcule begins expanding (Plate V, fig. 12), the coils rapidly unroll and are flung far out like a lasso thrown by an invisible arm. The retraction of the body down to the bottom of the cell is effected by means of a thread, which is seen affixed to the very tip of the inside wall, and looks—but only looks—like a continuation of the pedicle inside the envelope; from this basal point the thread passes up along the body and comes to an upper fixation quite near the base of the anterior flagellum; in fact, this posterior thread is nothing but a flagellum, which has temporarily lost its normal functions to acquire those of a retractile organ. The nucleus is spherical, and situated posteriorly, near one side of the body; it is rather pale, not very distinct, and shows a large central caryosome. The contractile vesicle is at the posterior extremity, behind the nucleus; sometimes a second one is to be seen, but though being able to empty and fill again from time to time like an ordinary contractile vesicle, it can hardly be considered as such, but is rather an accessory vacuole, whose function is to refill the true vesicle when empty.

I was not able to study the division inside the envelope, but on the 16th and 17th of February, I found, among a great number of empty cases, a good many small Flagellata which were rapidly swimming about, and whose appearance was that of individuals belonging to this species after leaving their shell; all characteristics were the same, except for the form of the body, that of a true, naked Flagellate, with two flagella, one of which was trailing behind (Plate V, fig. 13); it was certainly the retractile thread, which in this new flagellated state had recovered its true nature. It would have been interesting to notice the fixation of these small Flagellates and the construction of the shell, but my attempts in that direction were not successful.

The rather numerous species which make up the genus *Bicoeca* are all of very small size, and the distinctive characters are sometimes very hard to recognize; the form of the capsule, however, is very constant, and the length of the flagellum, and the position and number of the contractile vesicles, also furnish important facts. The taxonomy of the genus is however, anything but clear. *Bicoeca* and the construction of the shell, but my attempts in that direc- *lacustris* J. Clark, for instance, as indicated by Lemmermann (27), is considerably different from the same species as figured and described by S. Kent (this latter observer speaks also of a second very small anterior flagellum, which character would refer to *Stokesiella*), and at the same time both forms are not very far from our *Bicoeca exilis;* but the special shape of the capsule, as also the very exceptional length of the flagellum, seem to point to a special organism, which has not yet been described.

Histiona campanula sp. n. Plate VI, figs. 14–19.

During the autumn of 1917, I found in a pond at Pinchat, a small Flagellate which seems to belong to *Histiona,* a genus itself very little known. It was most generally found fixed on the threads of a certain *Zygnema,* very often forming long colonies of individuals, standing next to each other along the mucilaginous sheath of the alga.

The protoplasmic body, very clear and delicate, occupies less than half of the capsule (Plate VI, fig. 14), which latter is itself so thin and transparent that its structure is not determined without some difficulty. It may be compared to an inverted bell, broadly open upwards, and whose closed end would be stretched on one side, so as to make a horn-like prolongation; and owing to this special form the little case, which rests with its posterior point just on the surface of the mucilaginous sheath, appears bent down like a small flower (*Campanula*) on its stalk, the concave side of the capsule being towards the *Zygnema* (Plate VI, fig. 14). The little capsule is at the same time laterally compressed, and seen from behind looks somewhat like a sugar-loaf (Plate VI, fig. 15). The capsule is provided with a pedicle, thin and colorless, which goes right through the jelly and joins the surface of the cellulose membrane of the *Zygnema,* where it is most generally seen to attach by means of a shining little cushion. The pedicle, we must observe, is not a true pedicle in fact, but is rather a protoplasmic thread more solidly affixing the cell to the substratum; the jelly alone would not allow of a sufficient hold; and, by the way, we must add that the mucilaginous layer being generally perfectly invisible, the capsule seems to be provided with a stalk; as for the length of this stalk, it is of course equal to the thickness of the mucilaginous layer, about 13μ or sometimes a little more or less. The protoplasmic body, which appears as if suspended inside the capsule without anything to keep it in place, is also very individual in its appearance: rounded behind, cup-shaped anteriorly, it is drawn up on one of its borders (always the same, that which is in contact with the convex face of the shell) into a long trunk-like appendage, which passes beyond the capsule and protrudes into the surrounding liquid. From the tip of that peculiar appendage stretch right and left two thin and broad protoplasmic curtains, which, passing over the intervening space join at last the opposite border of the cell. In fact, these curtains, or veils, may be considered as representing the extreme borders of the cup-shaped body, which have grown very thin and transparent. One of these veils, moreover, is much more developed than the other, and about the middle of its course is drawn up in an angular lobe, to be affixed there to the inside of the wall of the capsule, and this in a very curious way, by means of a "nail," a very small bluish spot, which is never wanting. Very likely this very fixation of one of these curtains to the border of the cell is a result of the necessity for the cup (or anterior part of the body) to remain open, for the veil, if it passed straight over to join the border at the opposite side, would prevent small food-particles falling into the cup.[4] The flagella are perhaps more curious than any other element in this already somewhat paradoxal being. There are two: the first one, which we might call the principal flagellum, starts (or rather seems to start?) from one of the borders of the cup, which we may consider as the right one, and very soon bends to the left, describing thus the figure of an arc, and finally terminates freely somewhat to the left of that appendage which we called the trunk. This flagellum nearly always proves perfectly quiet, like a bow stretched over the cell as if for aprotection against the intrusion of any enemy. It is a genuine flagellum, however, and from time to time begins vibrating, but becomes quiet again after a moment. The second flagellum is perhaps even more curious: upon looking attentively at the left border of the cup, and more especially at the tip of the "trunk," we find there (Plate VI, fig. 16) two very small bluish granules, one behind the other, and from the upper granule is seen to start a rigid, straight thread, whose length nearly equals that of the body, the structure of the thread, which diminishes in breadth from the base to the summit, seems to be that of an axial filament covered with a thin layer of hyaline plasma. This thread immediately turns somewhat to the left, and there remains quiet, like a tactile organ or seta, from time to time, however, it is seen to move right or left, like the hand of a watch, or, very rarely, to swing in a few longitudinal undulations, but only to come quickly to rest again. Indeed it hardly looks, like a flagellum, but is such in fact, only it temporarily departs from its normal functions to acquire a new importance. If we look very attentively at the anterior small corpuscle of which we were just speaking, and not above but behind it, we can just distinguish a very fine median line or thread, which passes the posterior granule, then continues its way down towards the bottom of the cup-shaped body.

In short, *Histiona campanula* is in possession of two flagella, but both seem to have been considerably modified in either structure or function.[5]

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[4] I never could see the second of these two "curtains" affixed to the capsule, and this second curtain is in fact so much smaller than the first that it might be considered simply the normal border of the cup.

[5] These statements about the flagella are the result of a good many observations, but the animalcule is exceedingly small and delicate, and more than one doubt is left in my mind about some points of its structure. I have seen, for instance, on two different occasions a very short flagellum beating at the floor of the cup; was it a normal structure, or perhaps the basal part of the right flagellum, torn off from the affixing blue grannule?

It is not likely that these modified elements should be considered as the principal agents in the capture of food. Rather must we think that the large surface displayed by the cup with its velum-like borders has the function of a net; I have very often observed small nutritive particles to be thrown, or rather to get caught on the surface of the expanded cup; and suddenly, when it hardly touches the net, a vacuole is formed, inside which the microbe (most frequently microbes were the normal food) violently struggles for a time, then suddenly collapses, as if killed by some poisonous substance. The food-vacuole then glides along the velum and finally disappears inside the body.

The nucleus, spherical and with a big central caryosome, is very distinctly seen, in the hind-part of the body. There is only one contractile vesicle, not far from the nucleus; some other vacuoles, however, are at times visible, either containing food-particles or not.

Phenomena were not observed concerning the division of the body, but I have frequently met with individuals in the naked state, which either resulted from an act of division, or, in a majority of the cases at least, had left their envelope to take to a swimming existence. Their appearance is quite characteristic (Plate VI, figs. 17, 18), nearly that of a mandolin, whose handle would be represented by that peculiar "trunk" we have spoken of; but then, from this very extremity where the small blue granule is still to be seen, starts now, not a rigid thread, but a true flagellum, which, beating normally, draws the animalcule forward in a rapid course; the functions, indeed, of the thread have changed, and have become those of a normal flagellum.

As for the other flagellum, which we looked on formerly as the principal one, it is now seen to start from the posterior, slightly pointed extremity of the body, and immediately curves upward, describing an arc whose tip becomes lost behind the body, near the base of the trunk; this bow-like curvature seems at first sight to delineate the very contour of the body (Plate VI, fig. 17), which fact gives the little being a very peculiar appearance.

After swimming for a time, the little Flagellate comes to rest; it sets itself down on the surface of a *Zygnema*-thread, and firmly implants itself there by its posterior pointed extremity; and hardly is the contact effected when a small root begins piercing the jelly (Plate VI, fig. 19), growing longer and longer, till after a few minutes it reaches the solid wall.

I could not ascertain how the animalcule constructs the new envelope. It seems likely that the naked individual, when already fixed, can remain a very long time unprovided with any covering; I found once a colony where all the animalcules, about six in number, were naked, and three days afterwards, though living and apparently in good health, only one of them was provided with a capsule. It is interesting to note that the animalcule, as seen enclosed in the shell, does not occupy the same position as he did when free; the posterior point, from which arises the arcuated flagellum, must have become displaced upwards and to the right, at the same time that the body became free from the pedicle and climbed up towards the opening of the shell.

Salpingoeca polygonatum sp. n. Plate VI, figs. 20–28.

This small organism was found in the same locality as the last one, at Pinchat, on the long filaments of *Zygnema pectinatum,* and as a rule the threads were either abundantly provided with the little Flagellate, or had none at all.

The capsule (Plate VI, fig. 20–28) is about 17μ in length with a breadth of 4½ to 5μ only; it is nearly cylindrical for most of its length, but is somewhat broadened at the aperture, and is rather abruptly narrower behind,[6] terminating in a point which itself rests on a thin, very short stalk; but note that, as in *Histiona campanula,* the stalk is not a true pedicle, rather a root, which pierces the mucilaginous sheath of the *Zygnema* and becomes attached to the cellulose wall below. It may be observed at the same time that *Histiona* and *Salpingoeca* chose two different Zygnamas as a substratum, and that in the former of those animalcules the jelly coating was thicker than in the second; hence the longer stalk in *Histiona.* The protoplasmic body (Plate VI, fig. 20), cylindrical in shape, and about 10μ in length, looks as if hanging down inside the cell, its upper end reaching as far as the aperture, while the lower, rounded end is at some distance from the bottom. It remains entirely quiescent, as a rule, but from time to time the little organism shakes weakly from head to foot, or suddenly retracts to the very bottom of the shell. The nucleus, spherical with a big central caryosome, is in the anterior part of the body, normally between two layers of very small hyaline granules. The contractile vesicle, rather large, has its place in the posterior portion of the body. When the animalcule is seen somewhat retracted in the shell, but still with its cylindrical shape, it proves to be deprived temporarily of that special protoplasmic collar which is known in Craspedomonads, but instead, there is a thick layer of delicate purely ectoplasmic substance, which constitutes, in fact, the material for the building up of the collar. After a moment's observation, one notices the protoplasmic layer growing upwards, becoming cup-shaped and more and more lengthened, and at last it has grown into a funnel, whose length nearly equals that of the animalcule itself, but it is so thin and delicate that only the two borders, right and left, can be seen, and as for the anterior rim it remains (as in most of the Craspedomonadina) entirely invisible.

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[6] The form is nearly that of the flower in the genus *Polygonatum,* hence the specific name.

In *Salpingoeca polygonatum*, as well as in several other representatives of the group, one might easily believe in the presence of two concentric protoplasmic collars; but it would be a mistake, and I can only agree with the conclusions of Lemmermann (24: p. 564) relative to *Diplosigopsis frequentissima:* "As there is, in fact, in that species but one plasmatic collar, it must pass to the genus *Salpingoeca*." But when the same observer continues: "How it is with the other Craspedomonads where two plasmatic collars have been mentioned, remains to be investigated," I should go further yet and should not be astonished if both the genera *Diplosiga* and *Diplosigopsis* had to be abandoned.

The flagellum, about 1½ times as long as the animalecule, is rather thick and distinct, and points straight upward, swinging with longitudinal undulations that sometimes affect the whole length of the flagellum, and at other times only concern the tip.

I very often had the opportunity to observe the reproductive phenomena, which are very interesting here. In Craspedomonads, transverse division, quite an exceptional fact among Flagellata, has been several times mentioned, and S. Kent, for instance, figures it in *Salpingoeca gracilis*, on Plate VI of his great classical work. Within the capsule are seen the two products of the division, still united to each other by a very thin tubular film, which soon will cut in the middle and form the two new collars. My own observations quite confirm the facts; first the body retracts, then loses its shape and appears an ovoid mass, and finally this mass becomes transversally sectioned; the upper of the two animalcules thus formed then rapidly rises towards the aperture of the shell, drawing out at the same time an axial, varicose filament or future flagellum, which gets thinner and thinner, and forming also around the circumference a thin tubular protoplasmic film, which unites the two new individuals (Plate VI, fig. 21). Each of these then lengthens, and the upper one becomes somewhat curved, leans with its concave side against the border of the capsule, and disengages its flagellum (either by a simple cutting, or perhaps, as it sometimes seemed to be, by separation of two flagella, which existed before but were glued together); at the same moment, the plasmatic tube divides and soon retracts on each of the new individuals. As soon as the separation is achieved, the upper individual oscillates on the rim of the shell, turns slowly down, and its flagellum is then seen pointing forwards. The little organism contracts somewhat, and remains quiet for some time, then suddenly darts forward, carrying in front of it a very long flagellum, longer, it seems, that it will be afterwards (Plate VI, fig. 22). It swims very rapidly, describing large circles, then suddenly stops, rests for a time, starts and swims again, rests, and so on for probably a long time, but I could not determine the period. The length of the animalcule is 7μ. One sees easily the anteriorly placed nucleus, and behind a large contractile vesicle, sometimes with an accessory one; a short protoplasmic collar is also present. The entire process, from the moment when both individuals are distinctly separated but yet quite near each other, to the darting away of the little flagellated animalcule, has lasted but six or seven minutes. As for the lower individual, which remained in the cell, it expanded in a few minutes also.

But things are often quite different: the young animalcule, which has just disengaged from the cell, instead of darting forward, remains quiet for a time, as if hesitating, then, with a few very weak downward beatings of the flagellum, slips along the exterior wall of the shell it has just left, and soon settles quite near, lying along the mucilaginous covering of the *Zygnema* (Plate VI, fig. 24, *a*). However, it very soon stands up straight on the jelly (Plate VI, fig. 24, *b*), and ten minutes after the first contact with the substratum it is already seen to have produced a very short root (*c*), which soon will reach the cellulose wall (*d, e, f*). But during that time also, the first rudiments of the new envelope are deposited, a thin film around the body, along which the animalcule soon rises, leaving the empty posterior end of the capsule behind. In fig. 23, which is an enlargement of *f* in fig. 24, the body is seen to swell out laterally, so as to press everywhere on the anterior part of the cell and enlarge it, somewhat like a potter who moulds his clay, but instead of a finger, the whole body is here employed. A little later (Plate VI, fig. 24, *g*), the body has retracted a little, the widened borders of the capsule are free, and are even too much expanded but will soon contract a little. The animalcule, however, moves more and more upward, and finally shapes up the aperture of the cell, very likely in the same manner as will be described in the next species; but I could not follow the events to the end, owing to the animalcule being lost by accident.

But such a division into equal parts is not the only one that can be observed. I happened twice to meet with quite another process. Instead of an equal transverse division of the individual, a very small portion only was seen to detach; first a little bud (Plate VI, fig. 25, *a*) at the anterior end of the body, then (Plate VI, fig. 25, *b*), after complete separation, a spherical little being was formed, hardly 5μ in diameter, with a long flagellum and a normal contractile vesicle; then it lengthened into a little flagellate, whose small size was the only character to distinguish it from the normal individuals. I could not, however, follow the little swimming organisms very far, and cannot suggest any explanation of the phenomena. Perhaps the cases were purely pathological.

Encystment was often observed.[7] The body becomes a yellowish olivet green, then a golden yellow, and finally brown; a membrane has gradually been secreted, which grows finally very thick; at first it is transparent enough to allow of the contractile vesicle to be distinctly seen, but finally it looks quite opaque. A very curious fact which remains to be noted (Plate VI, fig. 26) is that up to the completion of the construction of the cyst, the anterior part is open, and from the opening are seen still protruding either the collar or both the collar and the flagellum. Only when the construction is entirely finished, these plasmatic appendages begin to retreat, disappearing in the cyst itself; this latter, then, is seen to be cylindrical, or somewhat swollen in the middle (Plate VI, fig. 27), and may be compared to a cannon shell, provided anteriorly with a circular opening, which also, however, will soon be shut. Old cysts, in fact, generally have no opening, yet sometimes an anterior aperture is visible, but in empty cysts only, whose contents have very likely been liberated.

The encystment takes place inside the capsule, yet sometimes cysts are met with which are completely free from any surrounding envelope, and, besides, are provided with a short yellowish stem attaching them to the jelly. The fact might perhaps be explained by the encystment of swimming naked specimens, which hardly arriving into contact with the *Zygnema,* encysted directly, rather than building their normal envelope. Plate VI, fig. 28, which also represents a cyst of *Salpingoeca polygonatum,* such as I have seen several times, formed during the night from specimens which had been seen expanded the evening before, shows yet another kind of cyst. The animalcules in these cases contract into a perfect spherule, yellowish and shining, and besides a thin shell has been constructed also, but balloon-like and without any opening at all.

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[7] Lemmermann (27), in his diagnosis of the genus *Salpingoeca*, speaks of the cysts in the following terms: "Cysts inside the capsule spherical or oval, with a thin membrane or naked." My observations on *Salpingoeca polygonatum* are such as to furnish us with new or different data.

A few words might be added here, about the taxonomy. In *Salpingoeca,* a very large genus and one which soon will become much larger still, the form of the capsule is very constant, and furnishes a very good—perhaps the only good—diagnostic character. Now I could find nowhere such a description as might apply to the species just described. It is very near to *Salp. gracilis* J. Clark, but the capsule in that species is not cylindrical, being rather drawn out into a long narrow point, which appearance I never could detect in *Salp. polygonatum.*

Salpingoeca lepidula sp. n. Plate VI, figs. 29–33; Plate VII, figs. 34–40.

As stated in the preceding chapter, it is sometimes very difficult to come to a precise determination of the species in the genus *Salpingoeca;* and only after much hesitation have I come to the conclusion that the present form ought to be described as new. It is, in fact, very nearly related to *Salp. buetschlii* Lemmermann, butin this latter, the larger size as well as the more rounded and swollen contour, and, more than all, the very considerable enlargement of the cup-like opening, do not allow identification with the present species. The affinity would be more evident with *Salp. amphoridium* S. Clark, and also *Salp. vaginicola* Stein, at least as Burck (6) figures them in his 1909 work,[8] but in these the form of the capsule is not the same, being more like *S. buetschlii* in general appearance, and having a narrow pedicle which is not present in *Salp. lepidula;* this latter, also, always possesses two contractile vacuoles.

However it may be, the species which we are now going to consider was found covering in great quantities the threads of a Nostoccacean, itself abundant in one of the aquaria of the Zoological Laboratory at Geneva.

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[8] In Pascher's "Süsswasser-Flora" (27), these two species are figured on p. 82, but with quite a different form from that given by Burck. It is quite possible, indeed, that Burck had studied new forms, which it would have been advantgeous to give as such.

The capsule (Plate VI, fig. 29), about 14μ in height, has the shape of an urn, elegantly lengthened, resting by a broad, truncated base on a very distinct flattened cushion, which itself is applied to the substratum. From that flattened base, the lateral curve first somewhat contracts, but very soon dilates regularly, up to a distance a little above the middle of the height of the shell; then contracts again, to finally dilate in an expanded border. The protoplasmic body, rounded behind and itself elegantly urn-shaped, fills only half of the shell; the nucleus is very distinct, as a bluish spherule about the anterior third of the body; two contractile vesicles are found, one right and the other left, one posterior and the other inferior, and they regularly alternate in their function, emptying about each third minute; one of them, somewhat larger, seems to be the principal one, the other being accessory. The flagellum, about 1½ times as long as the body, is most of the time visible only up to the middle of its length, on account of the rapid vibration of its anterior portion. The protoplasmic collar, which is particularly interesting, is mostly found with the appearance normal in Craspedomonadina, being like a funnel, whose height may be equal to that of the animalcule itself; but this protoplasmic funnel is subject to extraordinary deformations. The peculiar accumulation of very pure ectoplasm which generally fills the funnel all around the flagellum is seen, for instance, to move up along the funnel itself (Plate VII, fig. 40), or the very thin film of hyaline plasma which surrounds the plasmatic neck—perhaps the "Schleimhülle" or mucilaginous layer Burck speaks of—expands and covers the exterior surface; and the collar itself, now persisting, now collapsing or getting mixed with the invading ectoplasm, takes on the most diverse appearances. Plate VII, fig. 37, for instance, shows a shortened, thickened collar, whose border has especially thickened; in fig. 38, the same structures are represented, one minute later than in fig. 37; in fig. 39, at the same time that the ectoplasm expands on the inside of the wall of the collar, it climbs up along the flagellum; in fig. 40, the ectoplasmic film it seen left and right climbing upwards, many more figures might have been drawn, showing the most varied structure, for instance a film of plasma moving up on one side only and leaving the other free, or a big plasmatic spherule filling the entire opening of the capsule. Plate VII, fig. 36 gives an idea of a very peculiar process, which I was able once to follow quite at length: a big mass of ectoplasmic matter protruded from the mouth of the capsule, and became rounded, uniting with the material of the now retracted collar, and after a few minutes, from that protoplasmic spherule started lobes and thread-like protuberances, some of which got very thin and long, and at last reached down to the supporting alga and there became affixed. After a time, however, all the lobes and threads began retracting backwards. At 11 h., 10 min., the appearance was that shown by fig. 36, Plate VII; at 11h., 30 min., everything was beginning to re-enter the shell; at 11h., 35 min., the rudiments of a new funnel were seen to be expanding upwards; at 11 h., 45 min., the collar was normal again, and the flagellum, which had for a time disappeared from sight, extended freely in the axis of the funnel.

The overflowings of plasmatic matter, which are frequently met in this particular species, are very little known; in some forms similar facts have been mentioned, and Burck, who, however, did not happen to meet with any such cases, speaks of the subject in the following words: "I have not found in normal individuals such formation of pseudopodia as Sav. Kent says he sometimes met with. According to this observer, Choanoflagellata may even pass through amoeboid stages. Zacharias assigns some importance to the pseudopodia in the capture of food, and does not hesitate to call them organs of prehension. Francé also observed Lobopodia in several places on the body. In my opinion such structures occur only in wounded individuals, which are not far from death."

I do not think such occurrences must necessarily be associated with a state of disease, but could not help sometimes observing a certain coincidence between the development of these pseudopodia and the abundance of food, the animalcules capturing then with increased quickness very small special organisms (sulfureous algae?) which were extremely abundant all around. But this way of capturing food is certainly exceptional; the nutritive elements are mostly thrown down straight upon the plasmatic funnel, either on its outer surface or more often on the inner surface. When the prey are very small, they seem to be digested inside the wall of the collar itself; a small shining globule, for instance, such as was mostly the case, was seen to lose by and by the sharpness of its outline, then to swell up, turn pale, and at last disappear, without having moved at any time from the place it occupied. But when the prey are caught quite near the bottom of the funnel, a vacuole immediately forms around them, and they are quickly seen to disappear downwards. In one particular case, I saw a small shining particle fall on the naked neck of an animalcule that had not yet constructed more than half its shell; it was immediately caught in a vacuole, and slowly slipped down along the surface of the body, to stop after five minutes quite near the posterior extremity (Plate VI, fig. 33). The downward course seemed to have been effected in a spiral line, but I no more than Burck could detect the least sign of a fine spiral tracing, itself representing, as Francé will have it, the border of a spirally coiled funnel.

Burck (6: page 183) discusses in a few lines the evacuation of foecal material: "As regards defecation, my opinion is the same as that of Clark and Bütschli, who observed the elimination of digested food to take place very near the base of the flagellum inside the plasmatic collar." And further on he adds: "An evacuation of ingesta at any point of the body surface, as Fisch supposes it to occur, is certainly not correct." Though agreeing with these statements in general, I should propose a slight restriction concerning the last of these conclusions: if it must be considered certain that the evacuation of foeces is not possible at all points of the body indiscriminately, an exception ought to be made at least for the plasmatic funnel; I have often seen captured granules (which very likely were not to the taste of the animalcule) slowly glide *upwards*, and when near the very border of the funnel, be suddenly hurled away (as if by a sharp undulation of the border?), and curiously enough, these very small granules or microbes, which were probably provided with invisible flagella, after having proved quite inert and unmoved as long as they were included in the vacuole, when cast out at once began to toss about in a rapid dance, soon disappearing.

On the 9th and 10th of February, I met with a considerable number of small naked Flagellates (Plate VII, fig. 34), which could be referred without any hesitation to this same species; the size, the two contractile vesicles, nucleus, flagellum, protoplasmic collar, everything, in fact, indicated specimens which had left the shell and had adopted a free life. They swam quickly about, in small circles, without ever getting very far away; or they went straight forwards, for a distance hardly more than five or six times their own length, then stopped for a second or two, darted forwards again, and so on for a long time, proceeding by sudden intermittent advances. Perhaps some correlation might be supposed to exist between this sort of progression and a very curious fact, other examples of which are hardly to be expected in the whole series of the Flagellata, but which could easily be ascertained in each of the specimens observed, namely, that the flagellum, instead of *drawing* the body, was *pushing* it from behind; the flagellum, in fact, is posterior. But the small animalcule has itself changed in shape, it has shortened, and at the same time has taken the form of a top, pointed in front (in front when swimming, in reality it is the hind part of the body), and as for the plasmatic collar, it is seen reduced to one-quarter of its former length, but is rather thick and very distinct and still. The most interesting fact about that change in shape is perhaps the pointed extremity which never exists in the resting stage, and certainly seems an adaptation for swimming.

Plate VI, fig. 30 illustrates a case of division, nearly completed, such as I met with on the 8th of February. Very likely the process had been the same as that described in the previous chapter, for *Salpingoeca polygonatum*. The new Flagellate, ovoid in shape, is seen about to draw its flagellum from the shell it has just left, and beginning to glide down towards the supporting Alga. It is seen about ¾ of a minute later, nearly touching the ground but with its flagellum still attached to the border of the cup (Plate VI, fig. 31); one minute more, and the flagellum was free, pointing straight upwards, the protoplasmic collar had begun to develop, and the inferior extremity had become pointed. Very likely the rudiments of a new capsule were already beginning to appear; but at that very moment, the supporting *Nostoc* passed into some rubbish and was lost to sight. Other specimens, however, were found that same day and the next, at the stage just described (Plate VI, fig. 32), and they allowed of further observations:[9]

Immediately after fixation, the little body takes the form of a long top, with a distinct plasmatic collar, in which food particles are already caught; but the characteristic posterior point immediately changes into a very fine thread, and as the body gets higher and higher, leaving the ground behind, the thread gets longer, being just sufficient to prevent the escape of the Flagellate, but certainly unable to keep it upright. The body, in fact, is kept upright by a very thin envelope, which has already formed, the appearance being that of an egg in its cup (in Fig. 33, Plate VI, dotted lines indicate the rudiments of the shell). The new envelope is so thin, that one hardly can detect it, like two fine lines left and right of the point of fixation, and which diverge more and more to meet the body in its most swollen portion. The animalcule, in fact, rose while propping itself against the very borders of the partly constructed cell; but after a normal height has been reached the body ceases to get higher, the posterior thread disappears, the animalcule swells and moulds its body into the very form the shell will invest afterwards, and deposits all over its own surface a very thin pellicula, then retracts when the pellicula has become hard.

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[9] Several of these specimens seemed from their particularly large size to represent adults, which had left their shell without previously dividing.

The capsule is now nearly complete, and its walls soon will grow thicker; but one thing is wanting, namely the neck, or rather its widened border; and now—if two observations, both agreeing, suffice for the statement—the difficulty is removed in a very ingenious way. The animalcule lengthens somewhat, then bends first to one side (Plate VII, fig. 35), then to another side, leaving some material behind, acting in fact like the finger of a potter about to broaden the aperture of a cup, but here the displacements are exceedingly slow, very likely because, before leaving the contact portion of the border, the animalcule must wait till the deposited film is hardened.

From my observations, the construction of the capsule takes a long time, from morning to evening; but it is quite possible that in nature things go much quicker, owing to more favorable conditions.

Chrysamoeba radians Klebs. Plate VII, figs. 41–44.

Chrysamoeba radians, which Klebs described in 1892 (21), is not rare, and yet remains little-known; every observer has seen it, but very few have gone to the trouble of studying it at length. This study, it must be said, is little encouraging in itself, doubts and uncertainties are met with on many points, contractile vesicle, chromatophore, flagellum, division, capture of food, even the very existence of this Flagellate as a distinct species has been doubted, and so many little organisms are met with, whose study looks more profitable, that this particular one is easily neglected.

Scherffel, in 1901 (29), treats in a few pages of this little organism, and says in regard to the flagellum: "Besides the pseudopodia a swinging flagellum is sometimes seen, probably only when the amoeba is about to change to the Flagellate state, but as a rule there is none." A little further on, he says concerning animal feeding: "Klebs was not able to observe the capture of foreign bodies, and Senn says, in 1900, holozoic nutrition has not yet been observed. Yet in 1890 I had observed in a vacuole a little rod with the appearance of a Bacterium. This year in the spring I was able to verify the direct capture of green Algae and the evacuation of brown undigested remnants of food." Later on, in 1911 (30) the same observer, in his chapter under the title "*Chrysamoeba* and *Chromulina nebulosa*," seems to doubt his former observations about a flagellate stage, thinking the observed Flagellates might not have belonged to *Chrysamoeba*. "I consider more and more as probable the opinion that the genus *Chrysamoeba* is in fact a purely amoeba-like form, with never any flagella, comparable to those of organisms like *Chromulina* or even *Ochromonas*, but owing to this very absence of flagella, these forms are so characteristic in themselves that it seems advisable to retain *Chrysamoeba* as representing an independent genus."

Pascher, in the fascicle of his "Süsswasser-Flora" (27), which treats of the Chrysomonadina and is dated 1913, does not even mention the name *Chrysamoeba* in his list of accepted genera but only devotes a few lines to the subject, in a foot-note on page 13: "Klebs has created the name *Chrysamoeba* for *Chromulina*-like Chrysomonadina with two large chromatophores and an anterior contractile vesicle, which may turn into Rhizopods and develop pseudopodia after losing their flagella." In the same work, on page 90, Pascher comes again to the subject, and considers this same organism as belonging to the genus *Rhizochrysis*, "an artificially created genus that includes several Chrysomonadina of different origin, whose Flagellate-stage has been lost or in which, if present, it is not yet known." *Chrysamoeba*, such as Scherffel understood it, has now become *Rhizochrysis scherffelii* Pascher.[10]

During these last four years, while looking for Infusoria, I have often met with *Chrysamoeba*, in several localities and in all seasons of the year; and at the end of 1914 and the beginning of 1915, I devoted some time to its study (perhaps it is not quite useless to state that most of my collections of material were made after breaking a thick layer of ice), and was able to come to some interesting conclusions. The little organism I had to do with was, I must say, at the same time, *Chrysamoeba radians* Klebs, *Rhizochrysis scherffelii* Pascher, and also the *Chrysamoeba*-like *Chromulina* Scherffel speaks of. In my opinion, one single organism is here concerned, *Chrysamoeba radians* Klebs, a very distinct and characteristic species and genus.

Let us first insist on the fact that *Chrysamoeba* is a colonial organism. Scherffel, in 1908, sometimes found "large colonies of *Chrysamoeba*-like amoebae, where more than a hundred typical and well-developed specimens, all without any flagellum, were included in an amorphous, free-swimming jelly." At Pinchat, these colonies were very abundant, sometimes very much reduced and with only a few individuals, sometimes with 50 or more, and even 100; in one particular case, one of the colonies seemed to contain about 200 little amoebae; there are no limits, in fact, to the size of the colony, and as for the isolated specimens, which, it must be said, are always met with in greater numbers, they may very well be supposed to have been detached from the colony.

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[10] According to Pascher, the *Chrysamoeba* which Scherffel observed is not the same as that described by Klebs.

If we now look at one of these little *Chrysamoebae* (Plate VII, fig. 41), we shall recognize a small amoeba, 15 or 18μ in diameter, star-shaped or made more or less irregular by thin clear pseudopodia, finely granulated superficially, sometimes bifurcate or rarely branching, and whose length can be double the diameter of the body. These amoebae very slowly change in shape, and hardly in position, and the pseudopodia are seen to change also, retracting or reappearing, but without seemingly playing an important part in the locomotion of the animal.

The plasma, very clear, and often with many small vacuoles, contains a pale nucleus, whose structure is rather exceptional; most generally, a central caryosom is to be seen, around which is a narrow annular ring of light liquid substance, then an exterior ring of more compact matter; sometimes, however, only small vacuoles are seen in a homogenous grey nuclear substance. The nucleus is generally indistinct, hidden from view by the more evident chromatophore. This latter is in the form of a large golden-brown lamina, crescent-curved; it very often looks double, being first lengthened, then bent in its middle so as to acquire the form of the letter *V*. The contractile vesicle, very small and in an eccentric position, often can hardly be distinguished; its activity is seen to be very slow, and some patience is needed to become sure of its periodic emptying and filling, but I could ascertain that it functions normally, though its reappearance after closing is very slow. Here and there are seen scattered in the clear plasma more or less numerous small shining granules, and sometimes a large rounded mass of leucosine is found, of a very pure bluish grey. There is no stigma, and as for a flagellum, only in very rare instances is one to be seen, extremely thin and very slowly undulating; and its occurrence is always a sign that the amoeba is beginning to prepare for its swimming Flagellate form.

It is rather curious that the question of the capture of food should have been so long obscure, and still is so. Klebs could not ascertain its reality, nor did Senn observe more about it. Scherffel, however, as we said before, proved the fact, leaving no doubt at all. For my part, I observed in many instances the capture of small green particles, and more especially certain large (sulfureous?) two-segmented bacteria, which happened to come into contact with the amoeba and immediately were enclosed in a vacuole, soon disappearing inside the body, sometimes the pseudopodia were the means of capture, and the bacteria were seen to glide along them towards the central plasma. On two special occasions, diatoms were met with, nearly as long as the *Chrysamoeba* itself, and in course of digestion.

Such is the amoeba form of *Chrysamoeba radians;* but there is another form, that of a typical Flagellate, which Klebs had already seen and Scherffel afterwards described, only to doubt later his own observation and believe in a confusion with another organism. My observations, however, are such as to leave no obscurity. *Chrysamoeba* really passes through a flagellate state, or rather, one might say, voluntarily abandons one state for another. I have made, between December 29 and January 3, several experiments on the subject, which all gave the same results, five colonies of *Chrysamoeba*, nine, thirteen, twenty-four and fifty specimens in number respectively, were isolated on five excavated slides in clear pure water, and the amoebae were seen one after another to leave the jelly, which after two, three and four days was quite free of any tenant, while small Flagellata were swimming about. As an example, I may give here a textual copy of the brief notes taken for the smallest of these colonies:

Dec. 29. 12 o'clock. Nine fine specimens in their jelly; not one of them showing any flagellum; all are expanded as small star-like amoebae, 15 to 18μ in diameter.

4:30 o'clock. The same.

Dec. 30. 8:30 o'clock. Only four are left, a single one of them still in the star form; the others are rounded, and two of them quivering; on one of them I can distinguish the slowly undulating flagellum.

9 o'clock. The undulations are more rapid.

1 o'clock. Nothing is left in the jelly, and a small flagellated individual is swimming about.

On three or four different occasions, I was able to follow in one single animalcule, from beginning to end, the passage to the flagellated state. The amoeba slowly retracts its pseudopodia, then becomes spherical, and begins quivering from time to time, but without the flagellum, still too delicate, being yet visible; but it soon becomes distinct, and ten minutes after the first quivering of the spherule, it is seen rapidly to vibrate at the anterior pole of the now ovoid or pyriform animalcule; this latter then moves, leaves the jelly, and goes in a straight course, revolving on its long axis and swinging in a pendulum-like movement.

This *Chrysamoeba*-Flagellate (Plate VII, fig. 43), 18 to 20μ in length, is ovate, with the broader extremity either anterior or posterior according to the individuals, sometimes it is somewhat stretched behind. It contains a distinct, curved chromatophore, a small contractile vesicle near the anterior extremity, very small shining granules, and often a large spherule of pure leucosine; the nucleus generally remains indistinct, hidden in the curve of the golden-green chromatophore; the flagellum, very thin, hardly longer than the body, takes its origin from a very small notch, above which the anterior extremity rises in a small prominence.

After swimming about for a time whose duration I could not make sure of,[11] the Flagellate again comes to the amoeba stage, and contracts into a spherule (Plate VII, fig. 42), the surface of which soon begins showing small projecting denticulations, then each of these protuberances develops into a very thin pseudopod, which soon thickens, at the same time the flagellum gets more indistinct, it is thin, flabby, and remains for a long time at rest; now and then, however, it seems to come to life again, and oscillates for a brief moment; and finally it disappears, either collapsed, or rather—if a single observation entitles to ajudgment—resolves into a chaplet of tiny pearls, which are lost by and by.

Division has rarely been observed. As far as I know, Klebs and Scherffel alone have mentioned a few cases, and one cannot but be somewhat surprised at the rarity of the occurrence, when thinking of these big colonies whose constituent members might be supposed to arise from a single primary individual. As it is, I was able to verify a few cases of division, but only one of these could be studied at length. The animalcule was found already somewhat stretched in length (Plate VII, fig. 44, *a*), and was provided with two chromatophores, still united, which together had the shape of a *W;* to the left, a big mass of leucosine; to the right, a very large vacuole. Two minutes after that first observation, the chromatophores were separated, the vacuole had grown bigger still, and the leucosine globule had elongated (Plate VII, fig. 44, *b*); a little later, the 8-form of the body was much more apparent (Plate VII, fig. 44, *c*), the leucosine body was still more elongated; the big vacuole, after having acquired an immense size, burst and disappeared (Plate VII, fig. 44, *d*); the leucosine globule became divided into two parts, one of which passed to each of the new individuals just when the bridge between them was breaking (Plate VII, fig. 44, *e*); and the two new amoebae slowly went their way (Plate VII, fig. 44, *f*). The whole process, from *a* to *f*, had lasted twenty minutes. At no time had it been possible to detect the least indication of a flagellum. Scherffel, in the first case he studied, observed (1911, p. 315) that one of the new individuals, long before the separation, already possessed a slowly oscillating flagellum, but later on, when he again had the opportunity to observe a second case of division, he found that neither of the new individuals ever developed any. For my part, I consider it quite possible that in both cases Scherffel had seen rightly: in the first one, the amoeba immediately prepared for the flagellate stage, in the second it remained an amoeba.

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[11] The resting or amoeba state, whose duration is not difficult to notice, can last for days, but this duration may also be much reduced, and I have seen for instance a flagellated individual transform into an amoeba and develop pseudopods; but two hours later the little organism had disappeared again as if not satisfied with the new condition.

But if this case of division is the only one I could study at length, there is another which I should like to mention, which though not proving successful at last, is indeed hardly less interesting. The amoeba, when found, was already narrowed in the middle, and in possession of two distinct chromatophores, each of them in a *V* form, but united together by their adjacent extremities, like a *W;* so firmly united, in fact, that when but a narrow bridge alone separated the two new individuals, one of the chromatophores, which should have entered the new individual, had not been able to follow the movement, and remained in what we might call the "old" individual, while the "new" animalcule, quite deprived of any green matter, was just going away. A very narrow bridge, however, a mere protoplasmic thread, was still uniting the two amoebae; but danger was coming, an air-bubble approaching more and more, and to get rid of it I added some water to the side of the cover. The bubble retracted, but, as a consequence of a violent temporary shaking, the couple seemed to give up dividing, the bridge got wider, and by and by the new pale individual slowly "re-entered" the old one; after a moment a single amoeba was seen, with two chromatophores instead of one. But without that disastrous commotion, certainly an entire separation was quite near, and the new amoeba would have been colorless, just like the specimen which Scherffel once found, and figures on Plate VI of his 1901 work.

To conclude with *Chrysamoeba,* I ought perhaps to speak of the encystment. Some observations have been made which allowed curious conclusions, so curious, indeed, that . . . it would be better not to mention them, as possibly they relate to some other organism.

Chrysopyxis bipes Stein. Plate VIII, figs. 45–49.

"*Chrysopyxis bipes* has often been discussed in the literature, and yet seems to me not to be at all well understood in its organization." Such are the words with which, in 1911 (30), Scherffel begins his chapter about *Chrysopyxis,* and though the following pages of the work threw a good deal of light on the most interesting points, it will not be quite useless to report some further observations here. But before relating my own experiences, I should like to reproduce the lines with which Scherffel introduces the subject:

"According to the existing descriptions, *Chrysopyxis* is a flagellated Chrysomonad, provided with a shell. According to Stein, the animalcule is in possession of two flagella. Other observers, like Lemmermann, and Pascher, are of the opinion that there is only one flagellum. Iwanoff (1899) tried to explain these different statements by the fact that normally a single flagellum exists, but that this under special circumstances appears ravelled out in threads, so as to look branched. The two flagella of Stein would thus be reduced to a single branched one. Pascher (1909) accepts Iwanoff's opinion, and gives in his Plate XI, fig. 2*b*, a hypothetical drawing of a specimen in which the individual threads of the branched flagellum are wound up together at the base, and so explain the simple, thick, basal part of the flagellum. I had myself a good many occasions to observe *Chrysopyxis* in life, but I never succeeded in distinguishing an undulating flagellum, nor that "ravelling into threads" (Zerfaserung) which Iwanoff and Pascher speak of, and the existence of which I must confess to being very skeptical about."

Chrysopyxis is often met with, but my observations upon that interesting little Chrysomonad nearly all refer to a single station, that same shallow marsh at Pinchat where so many Protozoa have been found, and where in the spring of 1916 it was seen to cover in great numbers the long threads of *Zygnema.*

The capsule (Plate VII, fig. 45), about 13μ in length, is ovoid in shape, and at the same time pyriform, its anterior extremity being drawn into a very short tube, which, however, is hardly distinct enough to deserve the name. This little case stands upright on the vegetal filament, posteriorly drawn to one side in a very thin thread which turns down around the *Zygnema,* and describes a complete circle to rejoin the base of the cell on the opposite side.[12]

In most of the specimens, the capsule is quite colorless, but with age it grows yellow; it is mainly of a cellulose nature, as is easily shown by the chloride of zinc reaction. Iwanoff (19), however, speaks of the violet coloration as only in the shell, the annular ring remaining colorless. Yet I have seen that very ring distinctly colored, and probably Iwanoff's negative results were due to a defective quality of the reagent.

The protoplast itself, which hardly fills one-half of the capsule, contains a large olive-green or olive-golden chromatophore, in the form of a curved transverse lamina; then very small pure and shining globules, often a large spherule of leucosine, and, in the anterior part, a large vacuole, or sometimes two. Does this vacuole, or one of them, represent here a contractile vesicle? Iwanoff indicated it as such; but in one of the observed individuals I happened to see, at the posterior extremity, a very distinct vacuole suddenly collapsing, just like a normal contractile vesicle, and it may be asked if its true position would not be there, the anterior vacuole being concerned with the capture of food? A nucleus was not seen, being hidden, very likely, in the concavity of the green lamina. Iwanoff, however, mentions the existence of "a small nucleus, somewhat above the middle." As to the pseudopodia, which we must consider somewhat more at length, the spherical protoplast does not reach with its upper extremity so far as the aperture of the shell; it appears, in fact, suspended inside the cavity, and held in place by means of a plasmatic stalk, which arising from the median upper point of the body, goes straight to the mouth of the shell. But there, at the very opening, this stalk abruptly divides into four, five, six, and up to eight lateral threads, which diverge in all directions; they are very thin, smooth and rigid, genuine pseudopodia, in fact, and they behave as such; tiny microbes are caught, slowly glide towards the mouth of the shell, then reach the central stalk, along which they are seen to descend with a ten-fold rapidity. It often happens, when the fishing district proves rich, that the captured microbes come down in too great numbers, and are then obliged to accumulate, waiting for their turn; it is, indeed, a very interesting sight to notice them swallowed one after another down the main trunk (Plate VII, fig. 49).

I have never been able, among these rigid pseudopodia, to detect any appearance of a flagellum, nor any transformation of a rigid thread into a swinging one, and my own observations entirely confirm those of Scherffel (30), when he says: "Then also with *Chrysopyxis* animal-like ingestion of food occurs!" and when he adds:

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[12] According to Iwanoff (19), the construction of this ring is effected in a very curious manner: the animalcule, in the state of a naked zoospore, turns several times around the *Zygnema* thread, depositing behind itself a fine train of mucilaginous matter, which soon will harden into a complete ring.

"The protoplasmic body does not possess any flagellum, any more than does the amoeba stage of *Chrysamoeba.*"

"The protoplasmic body does not possess any flagellum, no more than does the amoeba stage of *Chrysamoeba.*"

But here also, as in *Chrysamoeba,* a flagellated state may appear, not only after an act of division,[13] but as the result of a total transformation, a process hitherto unknown in *Chrysopyxis,* but which I happened to verify on several occasions: the body becomes spherical, the pseudopodiar etract, and the little spherule begins quivering in its cell, a flagellum, in fact, is there already; then the spherule lengthens, reaches the aperture, becomes swollen anteriorly, so as to press on the tubular opening and force the exit (Plate VII, fig. 47), and finally leaves the cell. It looks now almost like a small flagellated *Chrysamoeba,* yet relatively broader and more rounded, with an anterior rapidly swinging flagellum, a normal chromatophore, small granules and often a leucosine body (Plate VII, fig. 48).

Both figs. 46 and 49, Plate VII, and especially the latter figure, represent cells which are entirely without the normal tubular extremity. A good many such specimens were met with, generally smaller than the type, about 11 or 12μ in length as well as in breadth. Were they young individuals? or was it a different species, perhaps Lauterborn's *Chrysopyxis stenostoma* (Süsserwasser-Flora," p. 28, fig. 43), which, however, is described as possessing two chromatophores. At any rate, it is well to reproduce here the words accompanying Pascher's diagnosis of *Chrysopyxis*: "a genus too little studied, and whose several forms are relatively little known."

Hyalobryon ramosum Lauterborn. Plate VIII, figs. 50–54.

In that same marsh at Pinchat was often found, in every season and often in great abundance, the Flagellate for which Lauterborn (23) created the name *Hyalobryon.* "The present genus of *Chrysomonadina,*" says the German observer, "deserves its name rightly to this extent, that it is positively difficult to distinguish clearly even the outlines of the colonies; as a rule one sees only the numerous golden-brown Flagellates, placed next to each other, without its being possible to decide how they are arranged."

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[13] Plate VII, fig. 46 shows such a case of division, which is well known today, whose process Pascher summarizes in the following words ("Süsswasser-Flora," p. 28): "Multiplication by longitudinal division of the protoplast, after which one of the parts leaves the shell in the form of a zoospore with a single flagellum." In this fig. 46, it is to be noted that, in spite of the process being already very far advanced towards its end, one pseudopod was still to be seen, relatively thick, and pointing straight up; microbes were still caught, and slowly glided along towards the body.

"*Hyalobryon* constitutes, like the well-known *Dinobryon*, tree-like or brush-like colonies, but which, different from the latter genus, do not swim freely about, but are fixed on water-plants or any other substratum. The number of the individuals united into a colony is very variable and sometimes goes to a hundred. Like *Dinobryon* the Flagellates inhabit special capsules, which here are of a tubular form and often are more or less, even strongly, bent; the basal extremity terminates in a short cone. Towards the anterior end, the tubes become somewhat narrower, and at their border look finely toothed. This apparent denticulation is due to the fact that the Flagellates, which always stand near the aperture of their capsule, periodically leave here a deposit in the shape of new rings of growth, on the inside of the somewhat enlarged border. As the process is often repeated, the tubes lengthen more and more; and many of them are seen, whose anterior part is formed of ten and more rings, resting cup-like in each other."

My observations are such as to confirm in a general way those of Lauterborn, but the accordance is not complete in some of the details. In the *Hyalobryon* I had the tubes were never narrowed near the summit; on the contrary, they were perfectly cylindrical up to very near their distal extremity, where they somewhat expanded (Plate VIII, fig. 50) instead of narrowing. Lauterborn's "Anwachsringe," or so-called "lines of growth," were mostly absent; sometimes, however, they were present, either on some of the members of the colony, or more rarely on nearly all of them, but they could in no case be considered as rings of growth; they were tubes, set in each other, each of them of some length (Plate VIII, fig. 54). There was even no difficulty in ascertaining the manner in which the construction had been accomplished; little Flagellates approaching had undoubtedly become attached just *inside* an empty tube, very near the opening and on one side only of the wall, then they grew up, forming first a whistle-like, then a regular, tube. Plate VIII, figure 54, shows this special arrangement of the new tubes inside the old ones, and at the top is seen, in the form of a simple sharp scale, the beginning of a tube whose construction was not continued, the Flagellate having, it must be supposed, left the place very soon after finding it.

It would then be necessary to modify the terms used by Lauterborn concerning the fixation of these Flagellates: "While in *Dinobryon* the younger cells are set *inside* the inner border of the old ones, in *Hyalobryon* it is the *outside* wall that is chosen as a point of fixation, and this point is very far from the aperture." Now there seems to be here some misunderstanding, Lauterborn is here speaking of the fixation as quite a different thing from the "lines of growth," which latter would in his opinion point to a single individual building a formal row of rings above each other, while according to my observations there are no "lines of growth" at all, but the animalcules would settle down in two different ways: either inside empty tubes, as we just described, or outside and generally very far away from the opening, these latter individuals would enlarge the colony in size and breadth, the former would only lengthen the existing tubes.

The second way, by simple juxtaposition, is by far the more frequent of the two (at least it was at Pinchat), and colonies are formed, whose appearance is very curious and variable, now it is a branching thicket, now the tubes seem to start from a single point (Plate VIII, figs. 51, 52), now they are parallel to each other, now straight or very often in a contorted bundle (Plate VIII, fig. 52).

The small inhabitant of the tube is indeed of a very timid nature, and leaves its tube at the first appearance of danger, the very transportation of a colony from the general collection to an excavated slide for purpose of better observation is enough to cause more than half its members to escape during the journey, and those which still are left also very soon take to flight. It is quite probable that the animalcules are just as ready to settle down as they are to leave, and empty bundles of tubes may very well get the preference, they then affix themselves either against or inside the tubes, but in this latter case quite near the opening, and according to the conditions the changes may be of such frequency that after a time a formal set of rings are seen one behind the other.

As for the tubes themselves, whose length may reach as much as 150μ (Lauterborn speaks of 55μ) for only 5 or 6μ in breadth, they are perfectly colorless, and "for an exact study it is necessary to have recourse to staining, which does not present any difficulty, as they very quickly absorb aniline colors" (Lauterborn). I may add that Dahlia tincture instantly colors the tubes a very fine peach-blossom tint, but this lasts only for a moment.

The Flagellate, 18μ in length and 3 or 4 in breadth, is spindle-shaped, with at its anterior extremity a short beak-like prolongation, and, behind, it is drawn out into a point, itself continued in a very thin thread, whose length may be nearly double that of the fusiform body (Lauterborn calculates it to 12μ; but I have seen it attain 30μ and more), and which attaches by its extremity far away on some point of the inner surface of the tube. This attaching filament is highly retractile, and from time to time draws the animalcule very far inside the tube. Its normal position is at some little distance from the opening. In the anterior part of the body is seen the chromatophore, a golden-brown lamina, but which only appears distinct on one of its sides, where it is seen double becauses of its recurving there on itself. Sometimes it looks broken in its middle, and the appearance is that of two rods joining each other at an angle. At its anterior extremity is a very small, but distinct stigma. In the same part of the body is a small contractile vesicle (Lauterborn found two), and behind the chromatophore lies generally a large, egg-shaped mass of leucosine; in the very pure and transparent plasma, tiny shining granules are disseminated; a very pale globular nucleus, with a small central caryosom, occupies the middle of the body, being mostly very indistinct on account of the chromatophore, in the folds of which it is lost. From the slight notch that is seen at the anterior extremity, two flagella arise; one of them, the principal one, about as long as the body, swings in rapid longitudinal vibrations, while the other, the accessory flagellum, only 5 or 6μ in length, remains motionless at an angle of 35° to the former.

A little further consideration is necessary as to the systematic position of this small organism. Lauterborn did not unhesitatingly propose this Flagellate as new. As far back as 1890, Imhof (18) had described, with the name of *Dinobryon buetschlii*—but in a few words only, and without any accompanying figure—a small Flagellate, very similar indeed to Lauterborn's *Hyalobryon,* but in this form the tubes are expressly stated to be set one within another, and nothing is said of the characteristic "rings of growth.[14]

The form I studied also looks rather different from Lauterborn's *Hyalobryon,* and this typical insertion of the tubes, in place of Lauterborn's "rings of growth," would seem to indicate another species, perhaps Imhof's *Dinobryon buetschlii?* But Imhof insists on the fact that in his *Dinobryon* "the anterior third part of the tube grows gradually narrower, down to one-third of the diameter next to the opening," while nothing of the sort is to be seen in *Hyalobryon* as I could verify in hundreds of specimens. Perhaps a third species is in question, or a particular variety, but the facts are not clear enough to allow any definite conclusion.

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[14] Stokes' *Epipyxis socialis* (34), which presents some likeness to *Hyalobryon,* seems to me, even more perhaps than to Lauterborn, very different from *Hyal. ramosum.*

Cryptomonas ovata Ehrenberg. Plate VIII, figures 55–57.

Pascher, on page 105 of the second fascicle of his "Süsswasser-Flora," accompanies the diagnosis of the genus *Cryptomonas* with the following reflections: "A very widely distributed genus, that sometimes forms "water-flowers" (Wasserblüten) and is frequently associated with Euglenae; is particularly frequent in water rich in organic substances and in sewer refuse. Nearly all of the forms are also planktonic. Only few of them are known, and an exact study would furnish with a swarm of species." In a note at the foot of the page, Pascher adds: "I hope to give within two years a detailed monograph of the freshwater Cryptomonadinae." To my knowledge, the work has not been published yet, and any information about this interesting, somewhat paradoxal little group, might have its importance.

Cryptomonas is indeed found everywhere, but most of my observations, dating from 1916, have been made on specimens from that same marsh at Pinchat of which mention has so often been made; later on, in January, 1917, a collection was studied from another station, Rouelbeau, where individuals were found at rest, enclosed in a gelatinous envelope, and often in course of division; and lastly, in December, 1918, a special form from a pond at Florissant, in the near neighborhood of Geneva, was studied again. In all these different stations, the species was the same—the same, at least, after determination by means of the keys in "Süsswasser-Flora"—namely, *Cryptomonas ovata;* but such was the difference between the forms of Pinchat and Rouelbeau on one side and Florissant on the other, that one might ask if it was right indeed to reunite finally Ehrenberg's two species *ovata* and *curvata* into one (*ovata*). The Pinchat-Rouelbeau type was in fact *Crypt. ovata* (with the variety *curvata* Lemmermann), and the Florissant type was the genuine *Crypt. curvata* Ehrenberg.

Ehrenberg, on page 41 of his classical work ("Infusionsthierchen"), gives the following diagnosis of *Cryptomonas ovata*: "Monad with an oval carapace, with compressed body, oval, large, twice as long as broad, $\frac{1}{24}$ millimeter in length, color green." Pascher (Süsswasserflora, p. 105), in his diagnosis of the genus, describes the general appearance in more precise terms: "Cell typically showing dorso-ventral differentiation, with generally a more flattened or even somewhat concave ventral side and often a highly convex dorsal side, which at the anterior end is not infrequently drawn out into a point. Anterior end diagonally truncated. . . ." In spite of all descriptions, however, it would be very difficult to recognize a *Cryptomonas* without a few explanatory figures, so much the more, that the form of the body is exceedingly variable, from that of an egg to that of a curved spindle. The size also varies within very wide limits, most of the specimens measure from 30 to 55μ in length, but some are very much smaller. At the end of September, for instance, a multitude of very small individuals suddenly appeared, which though certainly belonging to this species, measured only from 12 to 13μ (Plate VIII, fig. 56), they were nearly oval in form, but grew nearer and nearer the typical form, in the same proportion as the size augmented.

Let us now consider in its more intimate structure, such an individual as is shown by Plate VIII, fig. 55, which represents fairly well the typical *Cryptomonas ovata.* The organism is seen with its ventral side to the right, the dorsal convex one to the left, terminating in a point, or beak-like extremity, the posterior part of the body is rounded, the anterior diagonally truncated, and hollowed into a depression whose deepest part is nearer the ventral side.

The most exterior part of the body is represented by a colorless periplast, of a cellulose nature, in the substance of which extremely fine granulations are dispersed, whose significance, as we shall later see, is that of trichocysts. Inside the colorless investing layer are the chromatophores, olive-green as a rule, in the form of two curved laminae which are applied to nearly the whole inner face of the external layer, and joining each other by their respective borders with such precision that one might think of one chromatophore instead of two. They are, however, not very distinct, except on their right and left sides, on account of their being seen there through a greater thickness. The green laminae are paralleled on their internal face by a continuous sheet of grains of starch, sometimes so much pressed against each other as to deform into hexagona facets, but always making only a single layer. These starch grains must be regarded as a direct product of the chromatophores, and at the same time as a food-reserve, for they disappear in twenty-four hours with unfavorable conditions of life. Besides these amylaceous grains, others of quite another nature are found; large shining bodies, very generally two in number (sometimes one and rarely three) showing indistinct crystalline forms which might apparently be referred to the hexagonal system; they are always found in the same position, not far from the dorsal side, and just dorsal to the characteristic "cage" that will be spoken of later. These special bodies, which might be compared to the oxalate crystals of Rhizopods and Infusoria, are nearly always present, an interesting fact to note, in comparison with *Cryptomonas curvata* where they never exist. Very near the anterior extremity, and under the dorsal prominence, is a very distinct contractile vacuole, which collapses about every half minute, slowly to fill up again. The nucleus, lying in the posterior region, is round, very large, very clear, and has a small central caryosom. Around the nucleus is clear, pure protoplasm, in which are dispersed shining globules of an extremely small size, whose appearance is that of leucosine. The two flagella are seen to start from the base of that beak-like prolongation that terminates the dorsal side of the body, but they arise nearer the ventral than the dorsal side, that is to say, on the ventral wall of that deep cavity which has been erroneously called the pharynx. When the little organism is swimming, nothing can be seen of the flagella; at rest, one of them is generally seen rolled into a wide curve, on the ventral side of the body the other extending backward, on the dorsal side. In one single instance, and in a very much compressed individual, whose body was spread into a broad layer, I was able to detect a very small basal granule. The pharynx is a deep, tubular or, more exactly, fusiform *fossa*, which plunges into the body down to the middle of its length, and even sometimes lower. At its lower end, this cavity is conical; near the upper end it narrows to a tube, then again somewhat expands, and opens funnel-like to the exterior. It is now seen to be carpeted, all along its inner wall, by a continuous sheet of clear granules, in one single layer and in perfect setting, disposed in such a manner that the eye can divide the surface either in longitudinal or in diagonal parallel lines, in two series crossing each other (somewhat like the appearance of the shell in *Arcella* or, better, *Cyphoderia*.[15] In reality, these small granules or pearls are disposed along diagonal lines going from left to right and from above below, and constitute a kind of "cage" whose lower end reaches far down into the pit, and whose anterior end suddenly terminates in a broad truncature. The number of the diagonal rows may be counted (in big specimens at least) as sixteen, and each row comprises about the same number of pearls also, so that we arrive to a total number of 256 pearls. The cage, however, if my observations are right, is not absolutely continuous; it seems to be split on one of its sides, the dorsal one, and from end to end, one of the borders of the longitudinal fissure being somewhat incurved; the whole cage, in fact, might be compared to an enrolled leaf, whose edges do not completely join. The layer of pearls, as we said before, carpets the inside of the pit, but the actual form of the pit is not easy to determine; it is neither a tube nor a fusiform cavity. Could we suppose, for instance, liquid plaster to be cast into the fossa and then removed in the hardened state, it would perhaps give a figure somewhat in the shape of a very thick spoon without a handle.

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[15] Bělar (4) gives these grains, in *Chilomonas paramaecium*, as forming 8 longitudinal rows, with 10–16 grains for each row. In *Cryptomonas ovata*, which differs from *Chilomonas* only in the possession of chromatophores, the grains are more numerous, and the rows also.

In regard to the significance of this tessellated cage, Bêlar (4), in his chapter devoted to *Chilomonas paramaecium,* has studied the subject somewhat at length, and says: "I should not, as Nägler does, consider these granules as supporting elements (stützende Gebilde), but rather apply that term to the fibrillae on which they are set, and whose existence, if not clearly proved, may be inferred from the regular arrangement of the granules, and which in face-view could occasionally be seen indicated in Iron-Hematoxiline preparations. . . . Rather must these granules be considered as rudiments of the jelly-trichocysts (Schleimtrichocysten) of the Cryptomonads. . . . Künstler's and Ulehla's statement, that, after the crushing of the animalcule, threads come out of the pharynx, which had not been there before, I can confirm myself, a fact, however, which can but very rarely be observed."

My own observations are also such as to confirm the existence of the ejected threads, I have seen them on three different occasions twice in the living animalcule and once after the action of sublimate, but they are so very thin, so difficult to observe, that I did not, at the time, consider my observations as sure, and I should hardly dare to mention their existence, if my sketches, which date from a time when neither the works of Bêlar nor those of Künstler or Ulehla were known to me, were not in accordance with those of the Austrian observer.

After these considerations upon these special granules, Bêlar passes to a somewhat different subject, which we must also treat of here: "We might speak now of the superficial "trichocysts" of our Flagellate. Bütschli was the first to observe them, after him they were described by several authors, lastly by Alexieeff; Dangeard and Nägler do not mention them. They appear as more or less rigid, fine threads from 10 to 30μ in length, which cover the whole surface of the body. They were mostly found after treatment of the animalcules with acetic acid. I have examined living individuals, as well as others treated by acetic and other acids, but without succeeding, either with the simple microscope or by dark-ground illumination, in finding anything of the sort. Only after fixation with osmic acid and coloration by Löffler's method for flagella, could I distinguish these elements, but then they were found in every one of the individuals. They do not color intensely, but are sufficiently visible. Each of the animalcules seems to be surrounded by an assemblage of fine supple threads. These threads have received many explanations. Bütschli considered them as trichocysts, homologous to those of the Ciliata; Künstler as dead pseudopodia; Lanessan as cilia, which are rendered invisible by interposition of mucilage. In my opinion the first of these explanations has the more probability. These thread-like elements, indeed, are only found on dead animals, for if present in life they would not escape observation, owing to the obstacle they would profer to locomotion. We must then look for corpuscles in the body, which would explain the threads themselves. And indeed, in Iron-hematoxylin preparations siderophile granules are to be found in a moderate number, close under the pellicula, which might eventually be considered as trichocysts not yet exploded. As for an homology with the trichocysts of Ciliata there can naturally be none. The whole question, especially that of the significance of these elements, needs further study."

I have not been able to detect the presence of these special filaments, yet must confirm the presence of the siderophile granules of which Bêlar speaks, and with which we shall soon have to deal more at length. Before going, however, into that particular subject, I must now consider for a time the second form I spoke of, and which was found at Florissant, Ehrenberg's *Cryptomonas curvata.* The diagnosis, as given by this author, is the following: "Monad with a curved carapace, with a very much compressed body, large, twice as long as broad, $\frac{1}{24}$ millimeter in length, anteriorly and posteriorly curved somewhat in the form of the letter *S;* color green."

In short, the diagnosis is here nearly exactly the same as that of *Cryptomonas ovata;* the only difference is in the slight *S*-curve, and one can easily understand the two forms having been united into one, so much the more since *Cr. ovata,* with its special characters not found in *Cr. curvata,* is also sometimes in the form of an *S.* But there are, really, differentiating characters which we must now examine. The anterior curvature (towards the ventral side), and the posterior (towards the dorsal side) are not of great importance, yet it must be observed that the curvature was present in all the specimens at Florissant, though it was exceptional in the other localities. The ectoplasmic colorless pellicle was thicker, and the small globular bodies (siderophile granules) were more numerous, more distinct also, the chromatophores, curved behind, were more distinctly separated from each other. But there are two more distinguishing characters, of much greater importance: the layer of starch grains which lies upon the inner surface of the chromatophores in *Cryptomonas ovata* was quite absent in *Cr. curvata,* and the same relations obtained in the case of the two big crystalloid bodies, which never are present in these species; besides, a curious fact was seen in *curvata*: if from the bases of the flagella their line of direction was followed downwards, as far as the middle of the pharyngeal pit, one invariably found there a special granule, very distinct after compression of the animalcule, of a pale bluish hue, and which immediately reddened with borax-carmine, a kinetonucleus, very probably, which could not be detected in *Cr. ovata.*

After these considerations of a systematic nature, I come now back to phenomena which proved common to both species, and which, indeed, were the only inducement to my observing *Cryptomonas* at some length: I mean those sudden leaps or jumps, which have long been known in the genus, and those explanation has never been given, or at any rate has not been proved.

From time to time, without any apparent reason or on account of some discernible danger, the animalcule suddenly leaps backward, in an amazing jump, five, ten, twenty times its own length, describing an arc or even a nearly complete circle, and always toward the same side, its ventral or concave side facing towards the center of the described arc. When danger grows extreme, and the irritation is at its greatest, the jumps succeed each other without intermission for a few seconds, and at last the little Flagellate falls down exhausted, or sometimes bursts altogether, as if some inside rupture of equilibrium has been produced, which dislocates its different constituents, leaving the animalcule as a shapeless body, with nucleus, starch grains, chromatophores, etc., scattered here and there.

Where are we to find the cause of these sudden jerks, the organ that might produce them? The flagella are out of the question; they are too weak, and besides, those individuals that happen to be deprived of flagella jump just as well as the others. Ehrenberg looks for the explanation in a contraction of the "lateral trunk" the beak-like prolongation, if indeed such is the meaning of the only line he devotes to the subject: "Das rasche Anstossen des seitlichen Rüssels veranlasste offenbar das Hüpfen." But neither beak nor trunk are in the least degree susceptible of any displacement, Another explanation has been proposed, to which Pascher ("Süsswasserflora," p. 5) refers: "In some particular groups (Cryptomonadinae), springing movements are effected by sudden contractions."

This explanation, which seems to be commonly accepted today, but without anybody trying to prove the fact, has in my opinion hardly any probability. If we take a frog, for instance, and cut its four legs quite close to the body, how can we imagine it to make sudden leaps two and three yards distant, by means of a contraction of its body? Our *Cryptomonas*, after all, very much resembles a frog, and even supposing its whole body to be contractile, it would be difficult to explain in this way even a mere trifling displacement. Such general contractility in *Cryptomonas* is, besides, nothing but a supposition, I don't think any observer ever noticed any contractions, and for my part, I could find no sign of contractility at all. But even supposing the contractility to exist, the solution of the question would not be much advanced, I happened on several occasions to see the animalcule leaping, for one or two seconds at least, when (under the influence of a current of glycerine) the ectoplasmic layers of the body were already crumpled, inert and dead. *Cryptomonas*, in fact, jumps after death.

In short, the contraction theory remains a mere supposition, and I may feel entitled to propose another explanation, which, if not necessarily certain, seems based at least on real phenomena. But before developing the subject, I must state a fact, which does not seem to have been mentioned yet: beside these long leaps backward, other leaps may be seen to occur, even much more frequent, but of much less importance; the body becomes displaced to right or left, forward or backward, by very slight quiverings, and the same appearance is noticed as in Infusoria while they are discharging their trichocysts. Now we have seen that according to the researches of Bêlar and others, the very tiny granules that are found in the periplast of *Cryptomonas* may be considered as trichocysts; in fact, one might compare them also to the "slime trichocysts" which we shall later on speak of in *Trentonia flagellata;* and it can hardly be doubted, I think, that the slight quiverings in *Cryptomonas* are due to superficial trichocysts also.

My supposition, then, could be formulated in the following words: the sudden jumps backward are due to the explosion of big trichocysts, and these are represented by the granules which line the pharyngeal cavity.

There are, of course, several objections against this statement. For instance, after jumping, the animalcule seems not to have suffered any change in the state of the pharyngeal "cage." But that objection is not strong, in fact is perhaps in favor of the theory. After one jump, even two or three, the "cage" looks complete, but it would be practically impossible to detect the loss of one or two granules, and one of them, indeed, on account of its large size compared to one of the small superficial trichocysts, would suffice to cause a leap, its sudden transformation into a considerable mass of some light matter (fine granules, liquid material, slime, or gas?), which would find its way out through the narrowed aperture of the fossa, would certainly cause a violent reaction backward. But though after two or three of these leaps the pharyngeal cage is still intact, it appears, indeed, very different when frequent and violent leaps have rapidly succeeded one another; the cage, then, is seen to be disrupted, the granules, much diminished in number, are scattered in a shapeless mass; sometimes a very few persist, perhaps immature ones, which did not explode. Sometimes, when the animalcule is subjected to the greatest possible excitation, as when a chemical reagent, for instance, comes into play, the jumps rapidly succeed one another without intermission, then the body collapses, bursts, and in the remaining (more or less homogeneous) mass, no trace of the cage is left; as if the granules, not finding a sufficiently wide way of egress, had all exploded inside the body.

A second fact seems to argue for the same theory: the line of the leap, the figure described in space by the displacement of the body. It is always, as we said, an arc, or a circle when the commotion is particularly violent, and always—if I observed well—with the same orientation. Now the pharyngeal pit is narrowed in its anterior part into a tubular passage, somewhat curved towards the ventral side, and on the supposition of a sudden emission of matter through this aperture, the backward jumping must describe a circular figure with the concave side of the body facing towards the center. The very form of *Cryptomonas* might be supposed to necessitate that particular direction, but, as we saw before, *Cryptomonas* may present the most different forms, the smaller individuals being even oval in shape, and yet the jumps never show any difference at all.

But I must now point to a fact, which though appearing at first not to be related to this subject, might on the contrary prove of the greatest importance. At the end of January, 1917, in my collections from Rouelbeau, most of the specimens were found in the state of rest, enclosed in a thick gelatinous envelope, just like a larva in its cocoon.[16] Now, as often as I managed to cause a slight current of carmine-glycerine to come into contact with the cocoon, I could see the animalcules, violently distressed and yet unable to jump, revolve on their longitudinal axis, like a top, and at an extraordinary rate, perhaps twenty turns in a second, the whirling lasted for two or three seconds, and the animalcule collapsed—dead.

The only explanation of these phenomena seems to be, that the animalcule must have been submitted to a violent whirling current, in a circular or spiral direction, and the current could only take its origin from the inside of the cocoon itself, from the discharge, in fact, of some matter which only the animalcule itself could have produced. Now the direction of the tubular opening in *Cryptomonas* is such, that on the supposition of ejected material, there must have been produced the exact whirling current whose effect we have noticed.

As for the composition of the ejected matter, we remain in complete uncertainty. We shall see later on, that in *Trentonia flagellata* the spherules which fill the ectoplasm explode in a finely granular mucilaginous material, easily demonstrated as a white little cloud by using India ink. In *Cryptomonas*, the same result might have been expected, but some experiments with this method have proved fruitless. I distinctly detected, in some particular cases, displacements in the dark mass of infinitely small black grains, and which were certainly not due to oscillations of the flagella (which give quite a different appearance), but no white cloud was formed. The granules did not resolve in these cases into mucilage, but perhaps into very fine granules, mingled with liquid substance, whose effect in India ink would hardly be conspicuous.

Astasia mobilis (Rehberg) Alexieeff. Plate VIII, figures 58–61.

Under the name *Astasia mobilis*, Alexieeff (1) described in 1912 a Flagellate belonging to the group of the Euglenina, and which he had found in the intestine of a *Cyclops*. That Flagellate, however, had already been described by Rehberg in 1880, as *Lagenella mobilis*, but a genus *Lagenella* had been created in 1850 by Schmarda for another organism, and the name had to be abandoned here.

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[16] Some of the animalcules, inside the gelatinous pellet, were seen to divide longitudinally; but my observations on the subject are very few, and though confirming in the main those of Bêlar on the same subject, would not add any phenomena of importance.

Alexieeff, besides, pointed out that *Astasia* (*Lagenella*) *mobilis* is very nearly related to de Beauchamp's (2) *Astasia captiva*, found in a Rhabdocoele, *Catenula lemnae*, and perhaps might even be the same.

I have met at different times, in the intestine of several species of *Cyclops*—*prasinus* and others much bigger and which were not identified—an *Astasia* which easily corresponds to the two forms, *mobilis* and *captiva*, at least in its general characters, but was mostly of a much larger size. Alexieeff, and de Beauchamp both speak of 30–40μ, Rehberg indicates 102–103μ, but in the form I studied, while the very young individuals which we shall soon speak of were within these limits of size, normal adult or old ones, very likely much enlarged by accumulation of food, reached a much greater size, some, which in the spherical state generally shown when first isolated from the host, reached as much as 88μ in diameter, and after a moment, in their full state of elongation, developed to a length of 300μ.

Let us now examine one of these large individuals (Plate VIII, fig. 58):

As in all the other species of the group, the organism continually changes in form, its progression being effected by means of swellings and undulations passing from behind forwards. "The metabolic movement," says de Beauchamp, "is perfectly regular; the flow moving from behind forwards, like a large drop which glides along a wire, and there is nearly always a single wave, the posterior end hardly beginning to enlarge by the time the drop reaches the anterior end." These swellings are sometimes so broad, in comparison with the narrow interspaces, that the appearance of the animalcule becomes most singular, like that, for instance, of a tee-totum. The limiting periplast is colorless, thin, but very tough; striated all over with very fine and regular diagonal lines, about ½μ distant from each other. The body is nearly always entirely filled with grains of paramylum, in the shape of rectangles, but rounded at the ends, 3, 4, and up to 8μ in length, greenish-opalescent when examined singly; but when seen as a general mass they have a blackish appearance. These grains represent undoubtedly nutritive reserves, and diminish in size the longer the animalcule fasts. At the posterior extremity, these grains suddenly turn much smaller. In the plasma are also found a considerable number of tiny, shining granulations, colorless, 1μ in diameter. The nucleus (Plate VIII, figs. 58, 61), lying about in the middle of the body, is spherical, and entirely filled with extremely small granules, which are of variable size according to the individuals. When after compression the very tough nuclear membrane is suddenly rent, there is seen flowing out a large clear area of plasma in which all these tiny granules are dispersed. These instantly become red when treated with carmine stain. The contractile vacuole is often encircled by a crown of smaller vesicles, which after the systole will fill up the vacuole again. It is at the anterior part of he body, and empties into a large reservoir, which we must consider a little more at length. The fore-part of the body terminates in a slight prominence, a very small point, which itself borders the entranceof a very narrow tubular opening, this tube then plunges straight down, to open into a vast pouch or reservoir. But, fromthebottomof the reservoir, or rather a little higher up on one of the sides, are seen arising two very short threads, one of which, the longer, is directed upwards in a diagonal course and reaches up to near the top of the reservoir, whilst the other, much shorter, hardly reaches the middle.[17] These two filaments, which must be considered as two flagella that never extend any higher, each have a basal granule, and the two granules, very near each other, sometimes appear enclosed in a common layer of very pure, bluish plasma. This narrow bluish spot, however, is so indistinct that, doubting its reality, I should not have mentioned it here, if de Beauchamp, with whose observations I only became acquainted long after my own were made, had not said that after the action of reagents he found a small crescent-like black spot upon the wall of the principal vacuole.[18]

Such is the structure of the body in these big specimens of *Astasia.* But it may be added, that very large individuals, 80μ in diameter when in the spherical state, and 300μ when in full activity, are of exceptional occurrence; they are mostly so large when only one individual inhabits the intestine of the *Cyclops;* more generally there are two, four, eight or more individuals, in which cases they are smaller, and it must be considered as probable that they all came from one specimen, which divided and redivided inside the host. They are, in fact, commonly of the same size in the same *Cyclops* and, if we have in mind the extraordinary differences that are noticed between the individuals one happens to meet with outside the hosts, we cannot but recognize in this special uniformity in size the result of origin from the same parent organism.

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[17] The existence of rudimentary flagella does not seem to have been observed. Alexieeff simply says of his *Astasia* that it presents a flagellum or has none; and de Beauchamp that "sometimes a flagellum is present, but in most cases it is impossible to find any trace of one." Neither of these authors, however, has seen anything inside the reservoir, and de Beauchamp speaks of the flagellum as taking its origin—when present at all—from the entrance to the buccal tube.

[18] It is not impossible that Pascher considered de Beauchamp's black spot as a rudimentary stigma, for in the diagnosis of that species he says: "Stigma rudimentary, on the principal vacuole." But de Beauchamp certainly did not see it in this light, as he expressly declares he did not find any trace of a stigma.

Division is very frequent, and it seems curious that neither Alexieeff nor de Beauchamp were able to observe it; de Beauchamp states that when outside their host, the animals died in a few hours. Moroff (25), successfully cultivated his *Euglena quartana,* very nearly related to *Astasia,* in an organic medium(pea-soup, etc.), and declares that they thrive in purely inorganic cultures, but that after a very little while they perish.

For my part, I did not find any difficulty in keeping these organisms in very good health for three to five and even six days, after isolation in a drop of clear water on an excavated slide and under protection of a cover.

After creeping for a few hours. one of these big specimens contracts into a perfect spherule, and there lies unchanged, for a variable time, a whole day for instance; but if after having abandoned the animalcule in the evening we examine our slide early next morning, we find then not one spherule but two, sometimes already four, at 10 o'clock in the morning, those four are dividing again, constricted in the middle, at 12 o'clock, eight spherules are present, at 6 o'clock, sixteen are to be seen. On the following morning, the number will be 64, and finally in the evening, after the 8th division, one may count (approximately) as many as 256 small *Astasia,* some contracted, some elongated and lanceolate, many of them creeping or swimming round a lump of mucilaginous material where most of the individuals are still imbedded. Eight divisions have then successively taken place, after which the primitive single individual has been turned into 256 little Flagellates. This number, it must be said, must be considered as a maximum, which I never saw exceeded, but have observed to be reached four times in my eight experiments. At other times, however, long before attaining that number the young animalcules begin to detach from the common mass; we should also note that this mass itself is not always homogeneous, but may be very irregular in appearance. It is not a cyst, in fact, just as the little spherules in the mass are not spores; the jelly is more a fugitive secretion, which is easily destroyed, and perhaps it is only on account of the absolute quiescence after isolation on the excavated slide that the common mass remains compact to the end. But, if we take the mass of jelly and carefully compress it under the cover, these little spherules, born from division and destined to divide again, immediately begin to lengthen, creep about, and change their form, just like adults, and this may be made to occur at any of the phases of division, the second, third, fourth, etc.

The animalcules of the same generation are all alike, and of the same size but after the third (or fourth?) division, they differ from those which came from the first and second (perhaps also the third) division in an important regard: they have, or they may have, a flagellum.

In one of these small Flagellates, such as was born from the eighth division (Plate VIII, fig. 61), the vermiform body is very clear and still full of paramylum grains, but these are now very small; just at the posterior extremity two or three larger globules are seen (excretion granules), colorless and highly refractive. The hind-part of the body is itself pointed, sometimes drawn out into a glutinous thread-like tail. The nucleus is central, with the characteristic granulations. The contractile vesicle and reservoir are quite as we have already described, and from the bottom of the reservoir are seen to arise two flagella; but while one of these remains in a rudimentary state, just as it was in the big individuals, the other projects through the buccal tube to the outside, rapidly beating, and trailing the animalcule behind. This flagellum, however, is very slender, and comes off on the least excuse; many of the specimens do not have it, either having lost it already or never having possessed it.

According to Moroff (25), in his diagnosis of *Euglena quartana*, a very near relative to *Astasia*, division takes place as follows: "Division after loss of the flagella, either longitudinally into two new individuals, or into four parts after acquiring the globular condition." According to this observer, the second division[19] is at right angles to the first, and seems then to be transverse, the first being longitudinal; but if we keep in mind that all the indications of transverse division in Euglenae have been considered by Klebs and others as very doubtful, we must hesitate in accepting Moroff's statement. It is quite possible that even in the globular condition, after a first division, the two segments insensibly move or displace, and the next division, which is for the observer at right angles to the first, *i. e.*, transverse, is longitudinal in reality. In all circumstances where I could follow the process in its details, the division proved to be longitudinal; and in some individuals, which though already creeping about were yet in a state of division, one could foresee that the next division would be longitudinal also, this on account of the nucleus, which begins lengthening at right angles to the longitudinal axis of each of the new individuals, even before the separation of the couple is effected (Plate VIII, fig. 60), and which then will be divided along a line parallel to the line of the next division.

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[19] Moroff never obtained more than two divisions, that is, four young individuals.

In these young Flagellates, just beginning to divide, the anterior part of the body broadens, and at the same time hollows into a slight concavity, so as to take the form of a heart; the fissure gets deeper and deeper (Plate VIII, fig. 60), and in proportion as it lengthens downward, the lobes of the heart-shaped body become more and more separated, moving in a respectively right and left direction, with the result that at the moment the fissure reaches the end of the body, the lobes, or new individuals, have each of them turned 90 degrees, and lie with their posterior ends united only by a narrow bridge, the ultimate phase of the division, cutting right through the bridge, will then appear to be transverse, while in reality the division has been longitudinal. But this very narrow bridge, which at last consists only of the very tough periplast substance, is not easy to break, the animalcule, or rather now *both*, animalcules, struggle right or left, stretch and retract alternately, and above all, twist and turn on their longitudinal axis as they lie tail to tail; one of them, sometimes, is seen to twist to the left while the other twists to the right (in fact each twists toward its own right side), and at last the separating bridge, very thin and contorted to the utmost, breaks and liberates the two young Flagellates. The little *Astasia* is now free, and creeps or swims about with an activity more remarkable than that observed in the adult condition; no difference, however, is to be seen between those very young individuals and the old ones, except, as already said, in the normal possession of a flagellum.

Speaking of his *Euglena quartana*, a colorless species also, Moroff (25, p. 102) writes as follows: "I succeeded in obtaining individuals which showed with some distinctness a stigma. At the beginning none of the animals showed any trace of such an organ; but after a few weeks I was able to obtain some, in which the eye was apparent as a very small yellow spot, later on it had acquired an orange hue and looked larger, but later still the eye remained in the same condition, without my obtaining any further results."

Astasia mobilis never shows any stigma, yet I found once, not in a *Cyclops* but in a watch-glass where some *Cyclops* had been isolated, an *Astasia* (or *Euglena?*) seeming identical, at first sight, with the habitual parasite, but its paramylum grains were of a more distinctly greenish hue, and besides a small red stigma could be detected distinctly. On another occasion, I got from the intestine of a *Cyclops* an *Astasia* in which *two* small red eyes existed, one to the right and the other to the left on the anterior part of the body. This specimen, it must be added, was relatively broad and short, and perhaps not far from dividing.

That these two individuals belonged to *Astasia mobilis* is not certain. Alexieeff observes that in the *Euglena* group "transitional forms are found which lead from facultative to definitive parasitisms." Must *Astasia mobilis* be considered as a *Euglena* whose evolution terminated in parasitism, and in which individuals may be found, which retain some ancestral characters?

Euglena pseudomermis sp. n. Plate VIII, figures 62, 63.

This elegant little *Euglena*, which looks at first sight so much like a Nematode that one sometimes mistakes it for one, was found in two different localities, Pinchat and Rouelbeau, in every season of the year but always very rare.

The form is that of a needle (Plate VIII, fig. 62),[20] very straight, 165 to 190μ in length for only 7½ to 8μ in breadth, and it is quite near the anterior part of the body, on a level with the contractile vesicle, that the maximal breadth is found; from this region it diminishes in width by insensible gradation to the posterior extremity, whose thinness is quite extraordinary. Owing to a slight lateral compression, a transverse section of the needle would give a broadly elliptical figure. Seen along the narrower of the sides, it shows a characteristic swelling (Plate VIII, fig. 62), coinciding with the dilatation of the internal reservoir. The periplast looks shining and smooth, but in reality proves longitudinally striated, but the striae are so delicate and thin that only in very favorable conditions can their reality be ascertained. At the anterior end (Plate VIII, fig. 63), which is somewhat expanded, and in optical section appears to be provided with two tips, the periplast curves and invaginates towards the interior, but for a short distance only, and from this point is continued as a simply plasmatic film which lines the reservoir.

This characteristic pouch or reservoir, very long, broad and rounded at the base, becomes narrower towards the top, and at last joins the buccal opening. Into the hind part of the reservoir, and on one of its sides, empties at regular times the contractile vacuole, which is sometimes surrounded by very small accessory vesicles. A short flagellum is seen arising from the buccal funnel, a little behind the opening; it looks as if held in place by a basal granule, and at the same time appears to be lengthened posteriorly, like a hardly visible tracing, until it reaches a very distinct swelling about level with the middle of the reservoir. This swelling is caused by the presence, just outside the wall of the reservoir, of a large and very distinct globule, or kinetonucleus. Opposite the kinetonucleus, and on the other side of the reservoir, is the stigma, of a delicate brick-red tint; its form is that of a saucer, and seen from the side looks like a crescent whose convexity is turned towards the reservoir. Viewed from the dorsal side, it is seen to be a small roundish spot. In the immediate vicinity are sometimes seen very small red particles, which later on will join the principal mass. The internal protoplasm, very clear, and with myriads of extremely fine granules, is in its major portion occupied by a more or less considerable number of rod-like elements, which look like chromatophores, but without color. These are needle-shaped, of a very pure opalescent hue, generally 25 to 30μ in length for only 2 or 3μ in breadth, and often even much thinner, especially those which fill the posterior end of the body. They are set longitudinally, imbricating upon each other, and lie along the whole length of the body. They seem, in fact, to take the place of absent chromatophores, and if we keep in mind the statements of several observers that green chromatophores are susceptible, under certain influences,[21] of being reduced to the state of leucoplasts, it seems that in our *Euglena pseudomermis* also we are entitled to think of the structures as leucoplasts rather than as paramylum bodies or any other sort of pyrenoid. We might then consider this species as one of those very rare green Flagellates, which have lost their colored constituents, but which still keep the stigma, but only as a vestige of a preceding state.

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[20] In the figure, the animalcule is represented too broad relatively to its length.

Is this species, although deprived of true chromatophores, still absolutely holophytic, or would it to a certain extent take food from the outside in the form of microbes or particles of some other form? I have found in several instances small vegetal particles in the reservoir, and also inside the body protoplasm some small vacuoles have been met with, which contained little granules, like food-vacuoles in fact, but the explanation might be different, and the subject remains still obscure.

About in the middle of the body is seen the nucleus, whose appearance is most peculiar. It is generally found as a spherule, surrounded by a clear halo of lighter material, but this nucleus proper is itself a constituent part of a long, pale, grayish rod-like mass, which stains red with carmine, but much less easily than does the spherical nucleus. Sometimes, there are two of these cylindrical masses, with the nucleus in one of them only, or the cylindrical mass may contain, not one nucleus in its center (as is most generally the case, (Plate VIII, fig. 63), but several, regularly disposed, for instance three at equal distances, or four in two groups which are separated by a longer interval (Plate VIII, fig. 62).

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[21] Light would be the more important agent. According to Zumstein (1900) the chromatophores disappear as such in the dark, but persist as leucoplasts, and reform into chloroplasts again if brought back into the light. *Euglena pseudomermis* was always collected with a small net dipped into the black mud of the bottom of the ditch; it is quite possible that it lives in semi-darkness inside that "organic felt" which is found carpeting the ground.

Euglena pseudomermis slowly progresses forward in a straight line, then suddenly stops, and takes up its course again, slightly shaken by the beating of the flagellum. The little moving needle looks rigid, not deformable, but is not so in fact; from time to time it is seen to curve feebly, or become deformed by longitudinal undulations, very weak indeed but adding in a remarkable manner to the resemblance to a small Nematode.

The only two Euglenae which I could find in literature to have any similarity to *Euglena pseudomermis* are *Eugl. acus* Ehrenberg and *Eugl. acutissima* Lemmermann. But both are relatively much less lengthened, fusiform rather than like real needles; and besides, they have characteristic green chromatophores. I long hesitated as to the genus to which this little Flagellate might belong. *Menoidium* differs from *Euglena* in the complete absence of chromatophores. Would it have been better to make a *Menoidium* of this species?

Trentonia flagellata Stokes. Plate VIII, figs. 64–67 a, b.

In the analytical table which concerns the group of the *Chloromonadinae*, as given by Pascher (27, p. 177), the subdivision of the colored forms is into three genera, which are distinguished from each other as follows:

I. No highly refractive trichocyst-like corpuscles in the peripheral plasma layer.
  A. Cells changeable in form, more or less pyriform, narrower at the anterior end. *Vacuolaria.*
  B. Cells less changeable in form; at the anterior end diagonally truncate, long and narrowed posteriorly. *Trentonia.*
II. In the peripheral plasma, highly refracting trichocyst-like corpuscles. *Gonyostomum.*

Vacuolaria would then differ from *Trentonia* only in having a more pronounced change of form, and also by some difference in shape, the narrower part being anterior whilst in *Trentonia* it is posterior. As far as I could observe, there is no importance in the first of these distinctive characters, the changes of form in *Trentonia* being much greater than is generally thought, and as for the general shape of the body, if Cienkowsky's *Vacuolaria virescens* proves to be in accordance with the above-mentioned diagnosis, Senn's *Vacuolaria viridis,* which according to Pascher is intermediary between *Trentonia* and *Vacuolaria,* is in fact like *Trentonia* being described as inversely pyriform (verkehrt birnförmig), rapidly narrowed at the tail-like posterior end, strongly broadened and excavated anteriorly. On the same page 177, Pascher adds a note: "We must probably reunite with *Vacuolaria* Stein's *Coelomonas,* which differs from the former only in the possession of a single flagellum. The second flagellum, which lies very close to the body, probably escaped Stein's attention."

As for *Gonyostomum,* or more especially Diesing's *Gonyostomum semen* which is synonymous with Ehrenberg's *Rhaphidomonas semen,* it is nothing but a *Trentonia* again, but one in which are found, under the periplast, small highly refracting rod-like bodies which at times develop as cilia. Speaking of *Rhaphidomonas,* S. Kent (20) writes as follows: "Excepting for the presence of numerous and variously distributed trichocysts it closely resembles *Coelomonas,*" and Stokes (34) says of his *Trentonia flagellata*: "This is very similar to *Rhaphidomonas semen* (Ehrbg.) Stein, with two flagella and no apparent trichocysts. When first observed it was without hesitation identified with the above-mentioned European form." In the classical treatise of Engler & Prantl (12), one reads, page 171, concerning *Coelomonas*: "Doubtful genus. Like *Vacuolaria,* but with only one flagellum. Stein has probably missed the second flagellum. . . . The genus *Coelomonas* Stein must also very likely be abandoned."

Between all these Flagellates, *Vacuolaria viridis, Coelomonas grandis, Trentonia flagellata,* and even *Goniostomum semen,* the differences are so obscure, the general form of the body, the triangular reservoir, the chromatophores—everything, in fact, is so identical, that it is difficult to fancy anything but one and the same organism; in *Coelomonas,* the second flagellum, which in *Trentonia* is at the very limit of visibility, might not have been detected; in *Rhapidomonas,* the trichocysts have been exaggerated in their appearance. However it may be, it is as *Trentonia flagellata* Stokes (33), undoubtedly identical with what I have seen myself, that I shall consider the curious Flagellate we now come to speak of. It was found, rather numerous, in two different localities: Pinchat, where I found it in every season of the year, and Rouelbeau, where it was seen on one occasion only.

The most striking quality of this Flagellate is perhaps found in its special coloration, a beautiful bright green, which invests the whole body, and at once distinguishes it from the many other colored organisms that are swimming about. It is the pure green of chlorophyll, such as is found in the spherical granules of the higher plants, being without admixture of blue or brown.

The body (Plate VIII, fig. 64), measuring 40, 50, and up to 60µ and more, is in the form of a fish, two to three times as long as broad, somewhat compressed laterally, broader at the anterior end, narrower posteriorly, and often terminating in a point. The form of the whole body is slightly changeable, subject to alteration here or there, becoming now and then contorted or even excavated, but coming after a time to its original shape. The little green organism goes straight on its way, with a single flagellum visible, which is thin and straight, about as long as the body, and which hardly seems to move. From time to time, a sudden advance or leap is noticed, somewhat as in *Cryptomonas* but to a much shorter distance, and not always backwards but forwards just as well. To get an adequate understanding of the intimate structure of *Trentonia,* it is necessary to have recourse to compression, gentle, weak enough not to deform the body to more than a moderate degree, and yet strong enough to stop its movements (Plate VIII, fig. 65). The result required is, however, easily obtained, as the periplast is absent (or rather represented by a very thin hardened pellicula) and nothing prevents the body expanding. The body, in fact, may be considered as naked, so much so that with a stronger compression the animalcule is seen to transform into the shape of an *Amoeba,* displaying lobes or even extending into pseudopodia. Inside the very thin surface pellicle is a rather thick layer of very pure ectoplasm, of a peculiar appearance, opalescent and shining, and in it are seen dispersed a number of very small light vesicle, or vacuoles, which seem to be provided with a distinct wall. But in that external opalescent layer other elements are found, tiny spherical granules, very clear and brilliant, which often are seen to protrude over the surface of the pellicula, sometimes even in the shape of short setae.[22] They must probably be considered as trichocysts, the very structures which are represented by Stein in *Rhaphidomonas semen* as rod-like in form, but in the present species globular. Inside this ectoplasmic layer are the chromatophores. They consist entirely of small fusiform bodies, regularly disposed in two or three superimposed layers, with their long axes in a line parallel to the surface of the body. They are very delicate, and when compressed become rounded, showing at the same time a central lighter region. All together, these fusiform corpuscles make up a thick envelope, limiting a large clearer endoplasm, colorless, half-liquid in appearance, which contains the nucleus. This latter, very pale, consists of a homogeneous grayish plasma, with here and there a few very small nucleoles. At the anterior end of this colorless endoplasm, a large triangular space appears, the characteristic reservoir, into which from time to time is seen to empty the adjacent contractile vesicle; the reservoir itself communicates with the outside by means of a narrow channel, which opens not quite at the anterior extremity, but somewhat laterally.

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[22] In an animalcule that had been much compressed, along its longitudinal axis (Plate VIII, fig. 66), without any addition of a reagent, the whole surface was seen surrounded by very short radial filaments, which must have resulted from the exploding of these tiny bodies.

Of the two flagella, only one of them, the locomotor flagellum, can usually be detected. Both, however, have their origin in the small depression where the channel of the reservoir opens, and, so far as I could observe, start from the same spot, a little within the opening and on the ventral side of the tube. Stokes describes the locomotor flagellum as "apparently originating in the oval fossa; the other taking its origin on the ventral or lower surface a short distance behind the anterior extremity, and usually trailing, but I should not be surprised if there was a mistake in this statement. Further on the American observer continues: "The trailing flagellum is ordinarily extremely difficult to see. When the Infusorian is rendered uncomfortable and sluggish by prolonged confinement beneath the cover, or partially poisoned by iodine, then the vibratile flagellum, which is usually held stiffly in advance, the tip alone trembling, is flashed into sight as a rapidly undulating spiral, and the trailing appendage is also momentarily directed forward. . . . It is scarcely possible to believe that Stein would have failed to notice so important an appendage." My own observations entirely confirm Stokes's statements; but as for a mistake by Stein, I regard it indeed as quite possible. The trailing flagellum is so extraordinarily thin, and its refracting properties are so much like that of water, that it is only detected with the utmost difficulty. I studied *Trentonia* for many days without noticing any second flagellum, which, however, was there; afterwards, I was able to see it on many occasions. Some of my observations seem to prove that when the animalcule is at rest, the accessory flagellum lies stretched behind, and adheres with its tip to the substratum, keeping the body in place.

When describing the thick layer of opalescent ectoplasm which surrounds the body, I mentioned but two kinds of included elements, the tiny trichocysts and the small round vesicles; but we must now consider a new sort of inclusions. They are spherules, 2μ in diameter, of a very pale and pure bluish hue, and though extremely numerous, generally escape any notice, or rather are ascertained only after compression of the body; and the greater the compression the more numerous they are seen to be, they are in such numbers, in fact, that they seem to make up the whole of the plasma.

Singly examined (Plate VIII, fig. 67*a*), they are seen to possess a distinct membrane, very thin and smooth but suddenly, after a few seconds of observation, the membrane happens to burst, and through the opening expands a voluminous mass of mucilaginous material, mingled with a number of extraordinarily fine dust-like particles (Plate VIII, fig. 67). These globules, in fact, must be considered as quite a special form of trichocysts, the "Schleim-trichocysten" of German authors. But during all the time that the gradual compression of the animalcule is continued, there are seen to take place inside the compressed plasma small commotions, sudden disturbances of equilibrium, explanation of which is difficult to determine; but if India ink is used, the most external of the spherules, which are easily examined singly, are seen to eject a little white cloud when bursting, at the same time that a slight commotion is noticed in the adjacent lining ectoplasm; all these successive commotions that we saw occurring inside the animalcule must then also have their cause in exploding internal trichocysts.

India ink experiments enable us to observe still more curious phenomena: if the black current is made to touch an animalcule which is in perfect health, and is moving freely under the cover glass, the organism is seen to throw out here and there, now backwards, now forwards, or on one side, these same wh te little clouds which we observed before, and at each of these characteristic explosions, the animalcule leaps backward or forward or to one side. It is, indeed, a very interesting thing to follow one of these little animals while swimming at full speed, discharging without intermission its projectiles, like a man-of-war that would bombard the enemy. To these successive explosions we must attribute the characteristic leaps in *Trentonia*.[23]

I must mention, before concluding, the existence of gelatinous cysts, which I found numerous at Pinchat, in the month of May. They are very similar to those of *Cryptomonas ovata*, and contain either one single individual, or very often two, longitudinally applied to each other, undoubtedly a result of division. Sometimes, instead of two individuals, four are found, or two couples, one to the right and the other to the left inside the gelatinous cyst, which would certainly indicate a second division.

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[23] When discussing *Cryptomonas ovata*, we had recourse to the trichocyst theory for explaining the sudden backward jumps, and the facts here obtained may be of considerable value as indicating greater probability in the former suggestion.

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EXPLANATION OF PLATES V, VI, VII, VIII.

PLATE V.—Figs. 1–3.—*Pteridomonas scherffeli.*
1. Normal aspect; at the left side, captured food; at the lower end, the posterior crown of pearls.
2. Another specimen, with some "flagellopodia" vibrating.
3. Another, in course of division.

[NOTE—The flagella of the above figures have been strengthened slightly to insure reproduction.—EDITOR.]

Figs. 4–6.—*Dimorpha tetramastix.*
4. Heliozoan state.
5. Flagellate state.
6. Flagellate state, with some trailing pseudopodia.

Figs. 7–8.—*Dimorpha monomastix.*
7. Heliozoan state.
8. Flagellate state.

Figs. 9–13.—*Bicoeca exilis.*
9. Typical appearance.
10. Another individual, seen from the side.
11. Disposition of the flagellum in the enrolled state.
12. An animalcule retracted in the capsule, and about to throw off its flagellum.
13. Swimming form.

PLATE VI.—Figs. 14–19.—*Histiona campanula.*
14. Typical form.
15. Another specimen, seen from another viewpoint.
16. Extremity of the trunk-like appendage.
17. Swimming form, with anterior flagellum undulating.
18. Another individual, with posterior flagellum vibrating.
19. An animalcule in successive stages of the act of attaching after swimming.

Figs. 20–28.—*Salpingoeca polygonatum.*
20. Normal aspect.
21. Division, nearly completed.
22. The flagellated animalcule, as it comes from division.

23. An animalcule swelling up to construct the capsule.
24. *a* to *g*, a young individual, resting on the mucilaginous substratum, then straightening, pushing down a pedicle and beginning the construction of the capsule.
25. *a*, unequal division; *b*, the small flagellate which comes from the unequal division.
26. Encystment; the cyst is complete, except at the anterior extremity, where the protoplasmic collar and the flagellum are seen expanded.
27. Another cyst, entirely closed.
28. Another form of encystment, in a balloon-like capsule.

PLATE VI.—Figs. 29–33.—*Salpingoeca lepidula.*

29. Typical form.
30. Division; the young animalcule sliding down the old capsule.
31. The same animalcule, ½ minute later; nearly reaching the substratum.
32. A young animalcule attaching.
33. Construction of the capsule; above to the left is shown a large microbe which fell upon the neck, then slowly glided along the ectoplasm and stopped opposite the contractile vacuole.

PLATE VII.—Figs. 34–40.—*Salpingoeca lepidula.*

34. Swimming form of the animalcule, after abandoning the capsule.
35. An individual completing the construction of the capsule; the primitive fixation thread is still to be seen.
36. An animalcule, whose ectoplasm is expanding in filamentous pseudopodia.
37. Thickening of the protoplasmic collar, by means of intruding ectoplasmic material.
38. The same, a few minutes later.
39. Another specimen, with ectoplasmic material climbing up along both the collar and the flagellum.
40. An individual with a sheet of ectoplasm, climbing up along the inner surface of the collar.

Figs. 41–44.—*Chrysamoeba radians.*

41. The *Amoeba* state.
42. A flagellate, about to revert to the *Amoeba* state.
43. Flagellate state.
44. Division. In *a*, the division is as yet but little advanced; the two V-shaped chromatophores, together forming a *W*, are seen in the middle; to the left, a large mass of leucosine. In *b*, the body has lengthened; in *c*, it begins to be distinctly narrower in the middle; the leucosine mass has lengthened; the large vacuole is larger still; in *c*, the leucosine body is already rod-like in form; in *d*, the large vacuole has burst. In *e*, the separation is nearly effected; half of the leucosine body passes to each of the new individuals. In *f*, the new individuals are separated.

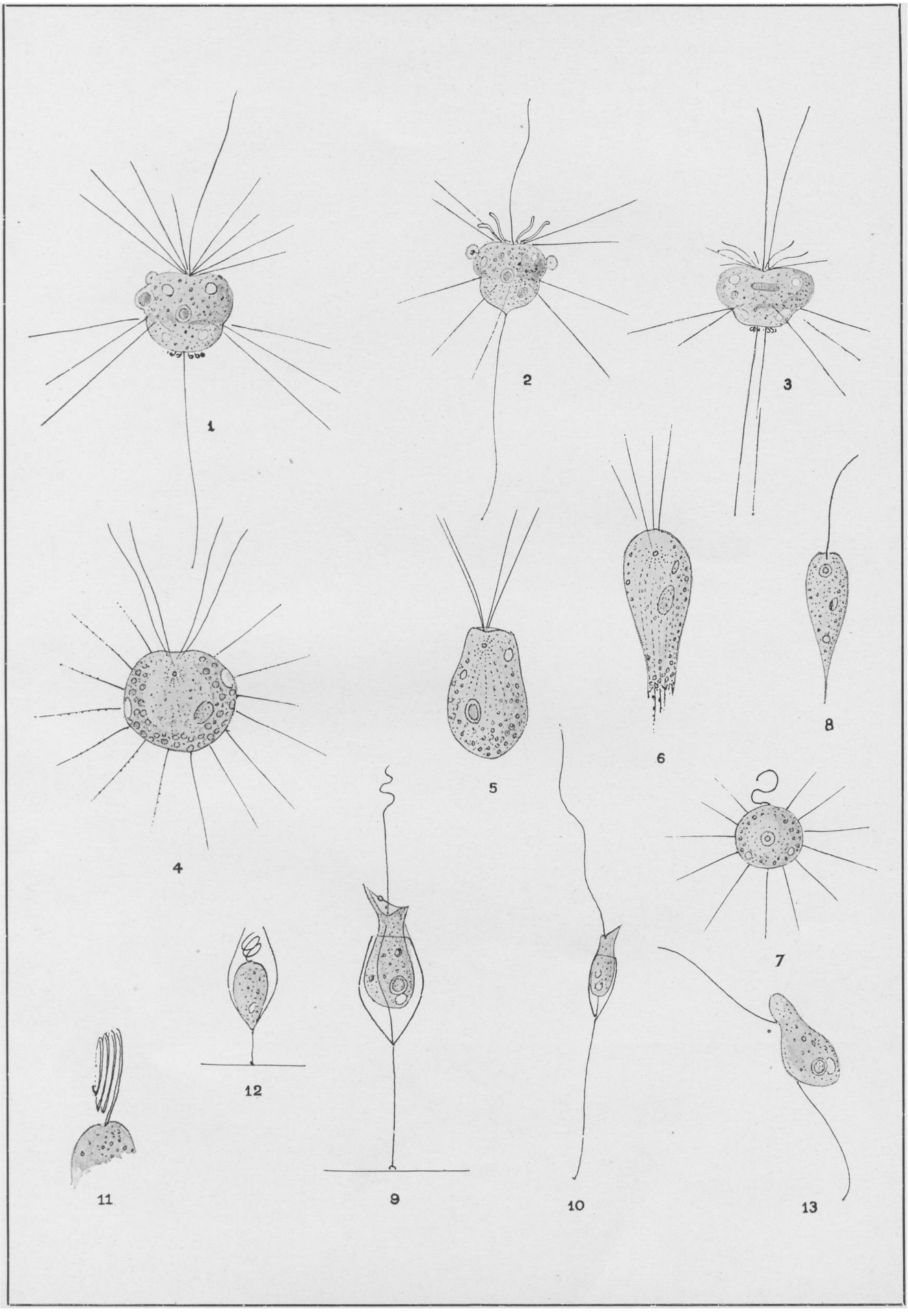
Figs. 45–49.—*Chrysopyxis bipes.*

45. Normal appearance.
46. Special form, without tubular opening; the body is in process of division; one pseudopod only, down which microbes are gliding.
47. An animalcule about to leave the capsule.
48. The same, swimming about after leaving the capsule.
49. No tubular opening. At the base of the psudopodia, an accumulation of microbes, about to disappear into the common trunk.

PLATE VIII.—Figs. 50–54.—*Hyalobryon ramosum.*

50. Normal aspect of the animalcule, with its caudal adhesive filament (which is represented too short, as well as the enveloping tube also).
51. A bunch of empty tubes, straight.
52. A bunch of empty tubes, contorted.
53. Tubes in a fascicle, attached to a diatom.
54. Several tubes inserted one within the other.

Figs. 55–57.—*Cryptomonas ovata.*
55. Normal appearance, with starch grains lining the chromatophores, and on the left the characteristic crystalloid bodies.
56. A very young specimen, ovate.
57. The "*curvata*" form, without starch or crystalloid bodies; on the right, near the pharyngeal fossa, is the kinetonucleus.
Figs. 58–61.—*Astasia mobilis.*
58. One of the large specimens. At the anterior end, the reservoir, with the two rudimentary flagella, and beside it, the contractile vacuole.
59. Nucleus.
60. Division.
61. A very young, flagellated individual.
Figs. 62, 63.—*Euglena pseudomermis.*
62. The animalcule seen from the side (represented too broad, relatively to the length).
63. Details of the anterior portion. Reservoir and kinetonucleus; to the right, the stigma; lower down, the contractile vesicle. Inside the plasma, leucoplasts, and the nucleus with its two cylindrical prolongations.
Figs. 64–67.—*Trentonia flagellata.*
64. Normal appearance.
65. Compressed body.
66. An animalcule, which was compressed along the longitudinal axis, showing radial setae (exploded trichocysts).
67, *a*. A mucilage-bearing spherule, in a small protoplasmic vesicle.
67, *b*. Exploded spherule, with a mucilaginous granular cloud.



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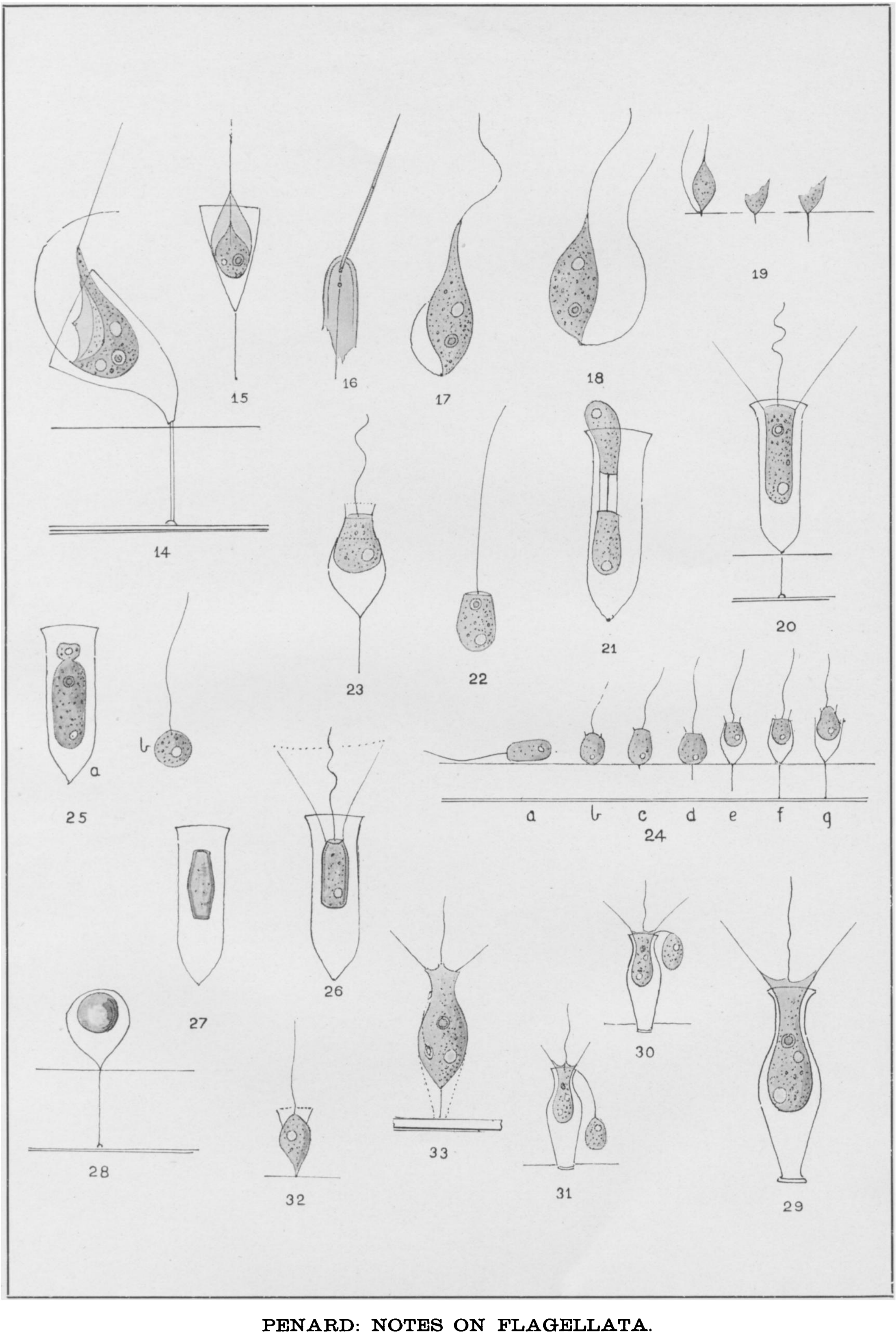

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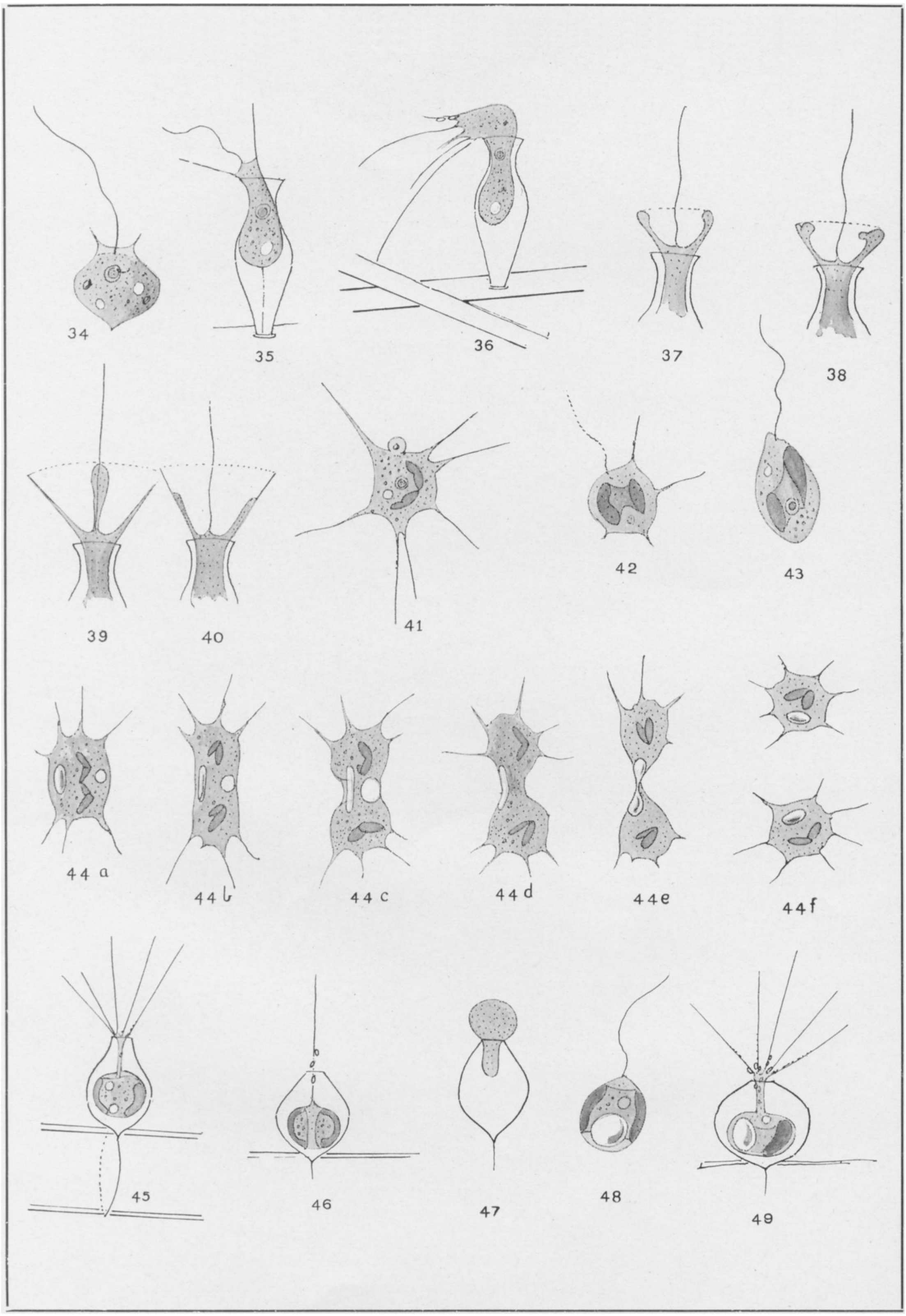

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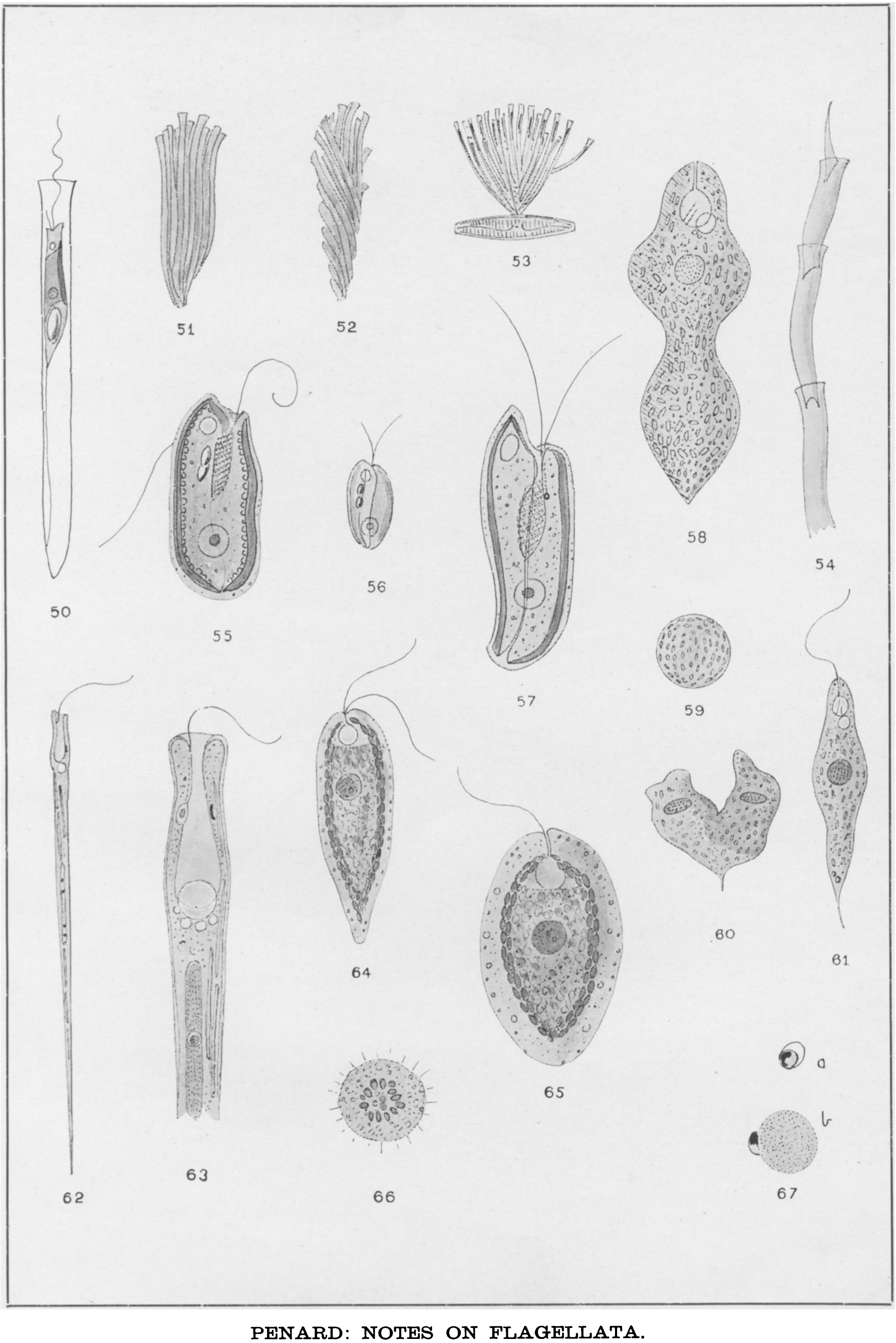

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